Curate Biosciences

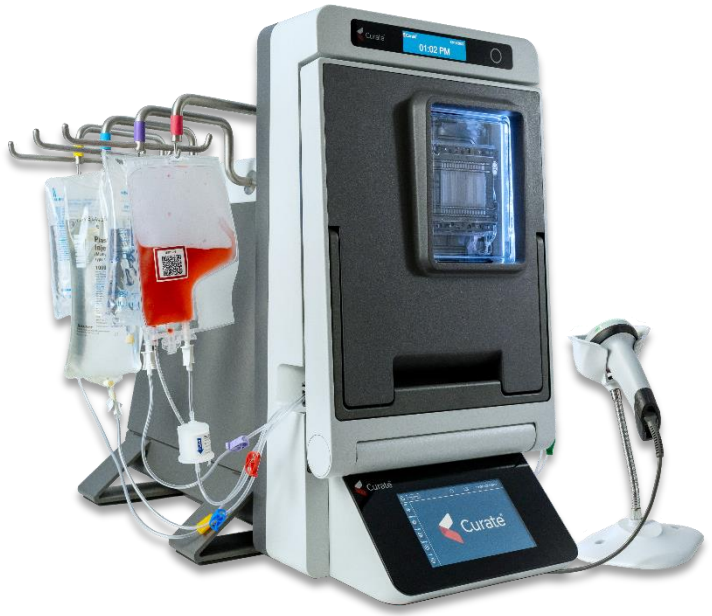
Advanced Cell Processing for Next-Generation Therapies

Tony Ward, CTO

Meeting on Mesa 2023

Curate Biosciences at a glance

- Privately Held, Located in Carlsbad, Ca.
  - VC and Biopharma backed
- Unique Centrifuge free cell processing platform
  - Consistent WBC recovery with unequalled purity and highest cell vitality
  - Available in US in 2023
- Interested in
  - Partnering
  - Co-development
  - Licensing/Tech
  - Regional Expansion (Asia)



Our Value Proposition:

The proprietary microfluidic Curate System will provide unparalleled starting cell product quality to enhance the cell therapy manufacturing process and improve outcomes for patients

Disclaimer

All statements and information in this presentation, other than statements of historical fact are forward-looking statements (within the meaning of the federal securities laws) based on assumptions concerning future conditions that ultimately may prove to be inaccurate. These forward-looking statements may be identified by words such as “believe”, “anticipate”, “contemplate” and “expect”. Many phases of our operations are subject to influences outside our control. Any one or any combination of factors could have a materially adverse effect on our business, financial condition and results of operations. These factors include competitive pressures, economic conditions, governmental regulation and policies and a variety of other conditions. Accordingly, if the estimates and assumptions underlying the information in this presentation are not achieved or proven to be invalid, actual results will differ from those projected, and the difference may be material.

We caution investors not to unduly rely on any forward-looking statements. The forward-looking statements speak only as of the date hereof. Curate Biosciences has no duty to update any of these forward-looking statements after the date hereof, nor to conform prior statements to actual results or revised expectations.

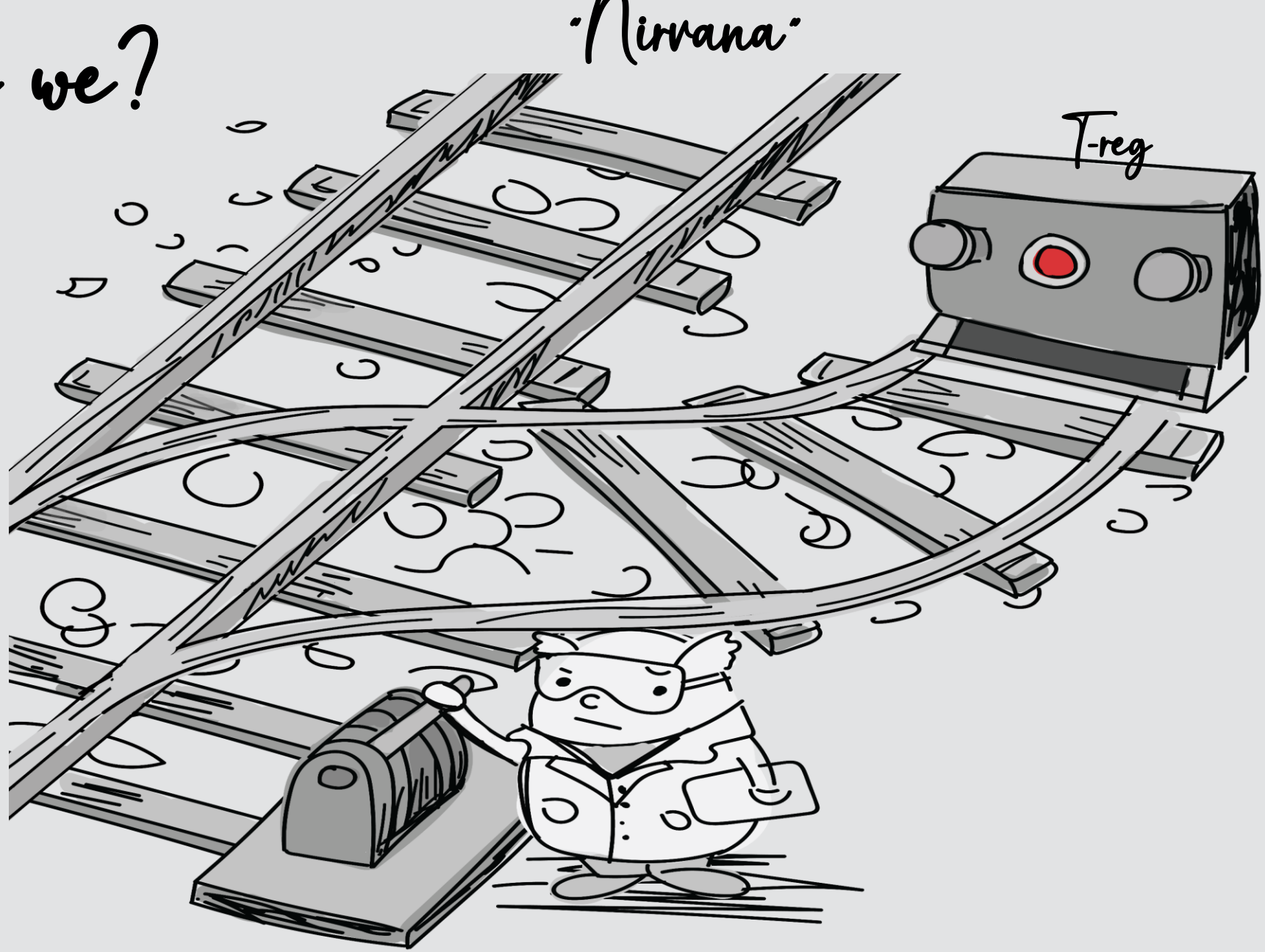
The products and techniques presented herein are not approved by the FDA to diagnose, treat, cure or prevent any disease.

What are the ways that Curate's microfluidic DCS processing improves process?

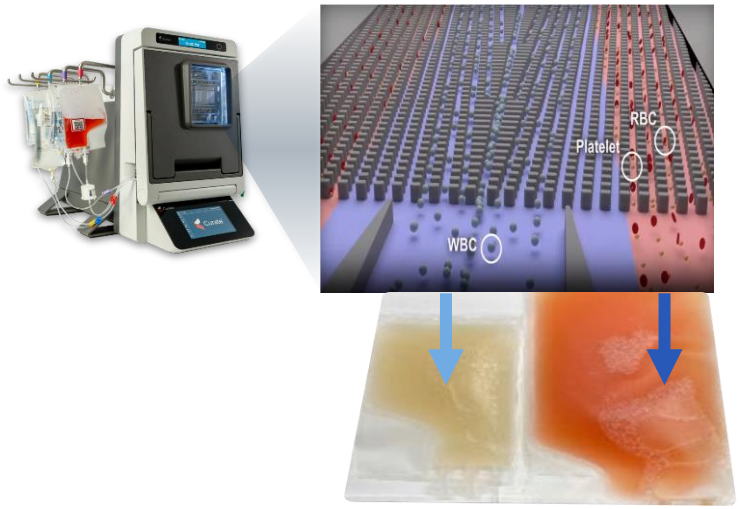
Key Objectives	Current Methods: ~>18% out of specification[1]	Curate Benefits
Optimal Recovery Esp. Younger T cells	Inconsistent Sub-optimal Recovery	Consistent Unbiased Recovery of all T subsets
Highest Purity	Variable, Poor Purity	Industry Leading Purity
Consistent Expansion & Yields	Inconsistent Yield[2]	Improved, Consistent Yield
Highly Engineerable	Inconsistent Engineerability	Highly Engineerable
Unperturbed	Perturbed Pelleting/Chemicals	Unperturbed

1. Legend CEO talking about Carvykti – Fierce Biotech Interview (12/22)
2. 20% of people are dying before they can get the CAR-T (Endpoints, MD-Anderson 3/23)

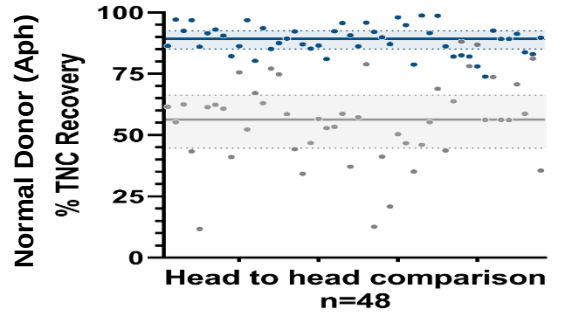
Where are we?



Microfluidic Deterministic Cell Separation™ (DCS) and its key benefits:

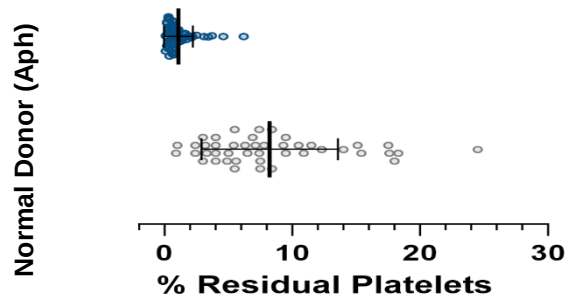


- Gentle, uniform processing with virtually no cell loss (Array efficiency: 96.5%)
- Same device separates, **washes**, concentrates individual cells in single process.
  - Processing rate of 50x10^9 WBC/Hr
  - **>3 Log wash in <1 second**
- **Requires no Ammonium chloride, Ficoll**
- **Does not pellet cells**

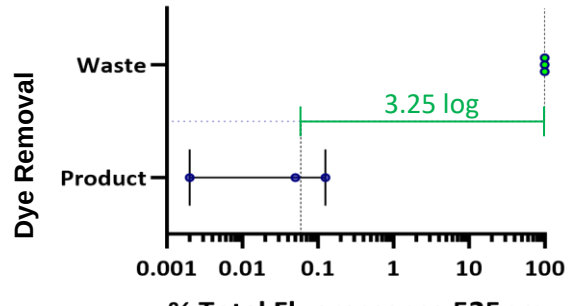


~2x Unbiased Recovery

DCS 89.3% 6.6 %CV
Ficoll 56.4% 30.2 %CV

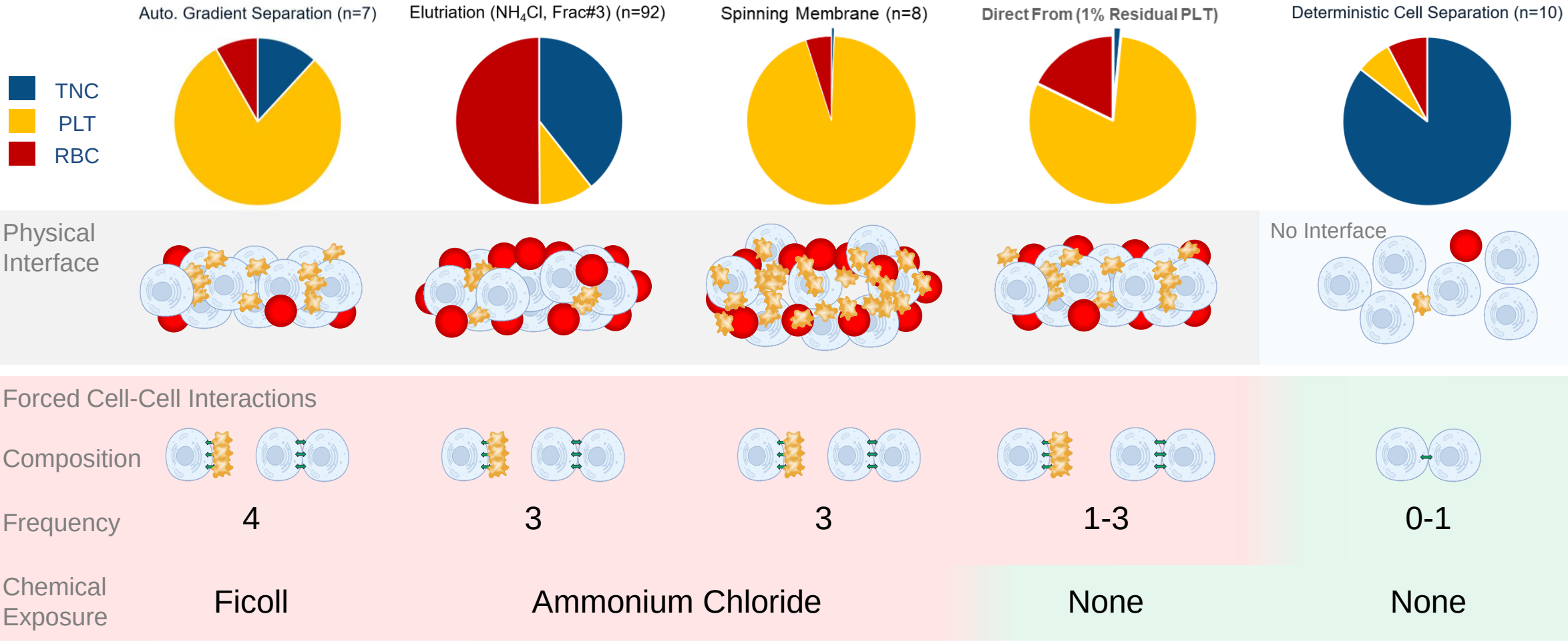


Up to 14x fewer platelets



>3 Log Wash

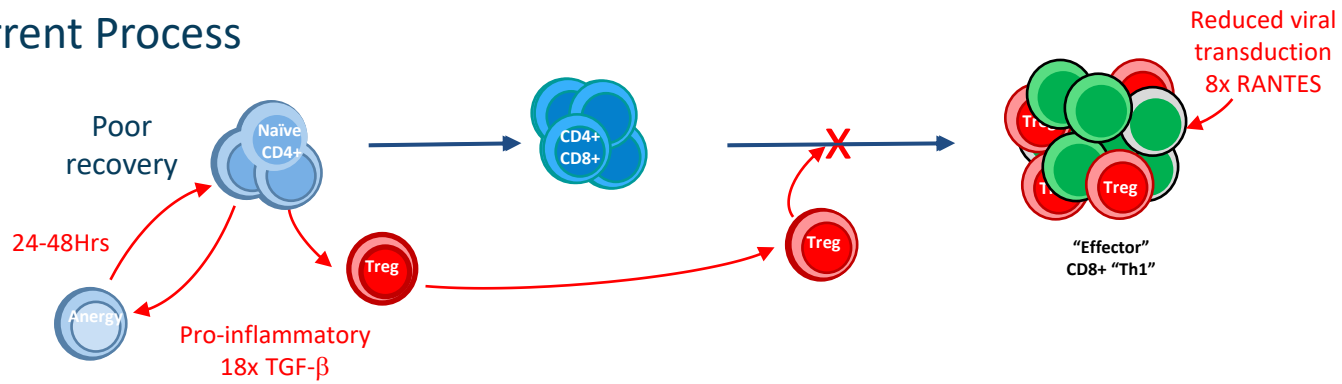
Legacy Technologies are highly variable and force cell-cell Interactions



Contamination Data is from non-Curate evaluations of each platform. Spinning Membrane is Mfg. data with platelet depletion program (~80% depletion) as presented by FHCRC at ISCT '2023, Direct from assumes 99% efficient plt removal

DCS Wash improves ***Pathway Specific*** Impact on Prototypic CAR-T Process:

Current Process

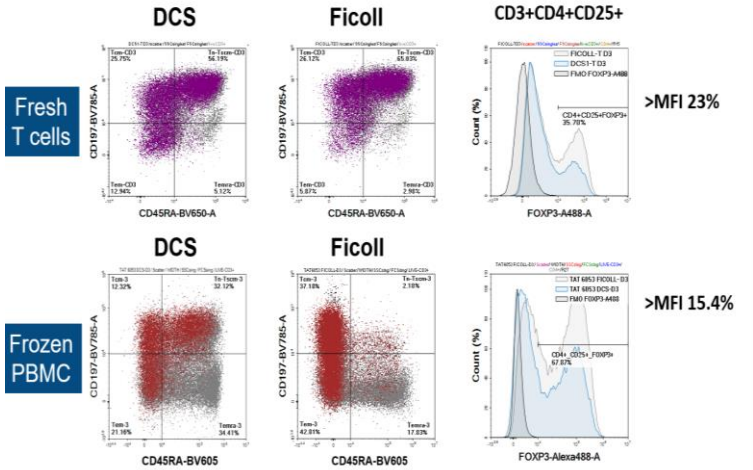


Naïve CD4 diverted to Treg/Anergy

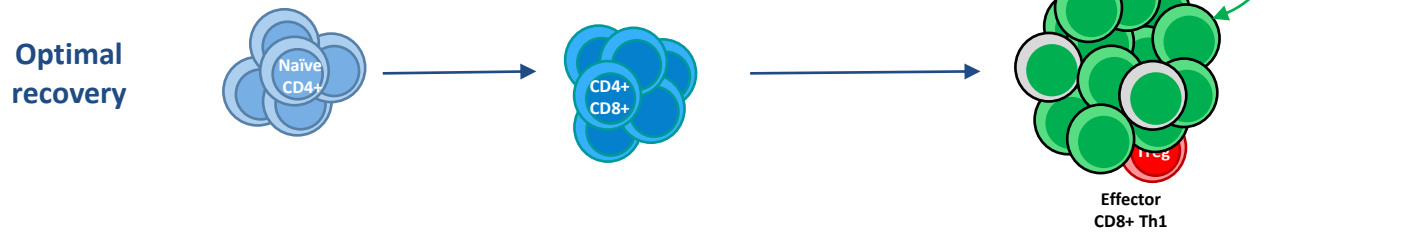
Incomplete conversion

Treg suppress expansion

Less CD8 effector per naïve CD4 T-cell



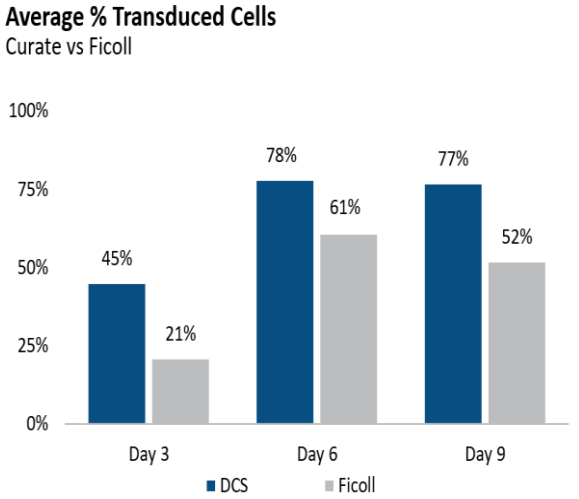
DCS Microfluidic Process



Address: Pro-inflammatory Anergy T-reg promoters

Conversion to CD8+

Unimpaired Expansion
1.3x Day 12
~50% fewer Treg

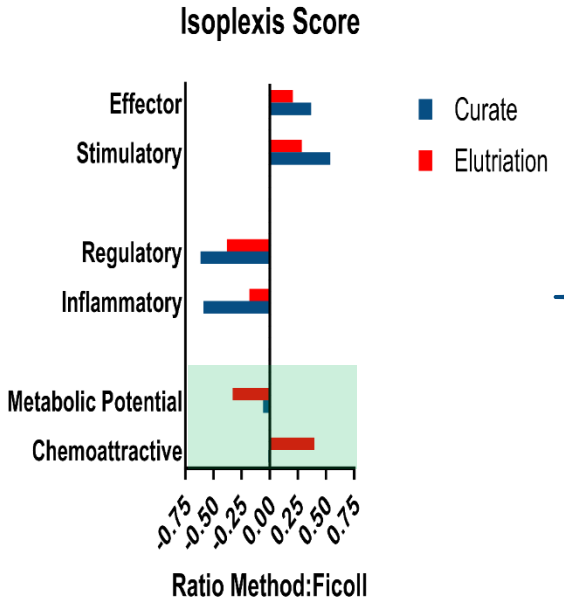


Detailed data may be found at: www.curatebio.com/publications/WhitePaper

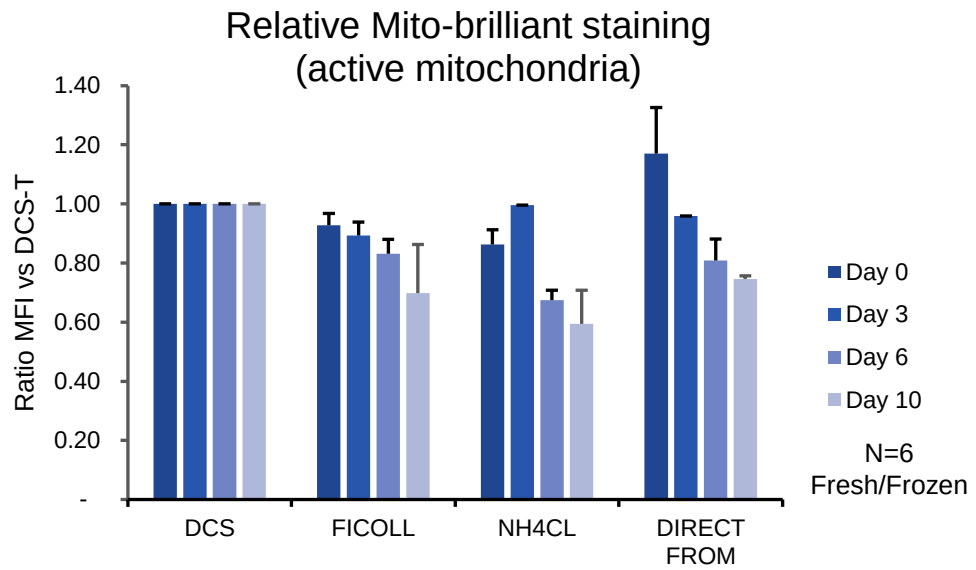
Elimination of lytic agents and centrifugal processes improves active mitochondrial mass in expanded T cells up to 40%

Fitness for Purpose

- Pathway Biology
- Avoidance of Chemicals
- Avoidance of cell-cell contact



3rd Party Data



T cells exposed to Ficoll, Ammonium Chloride or –ve selection directly from start matrix

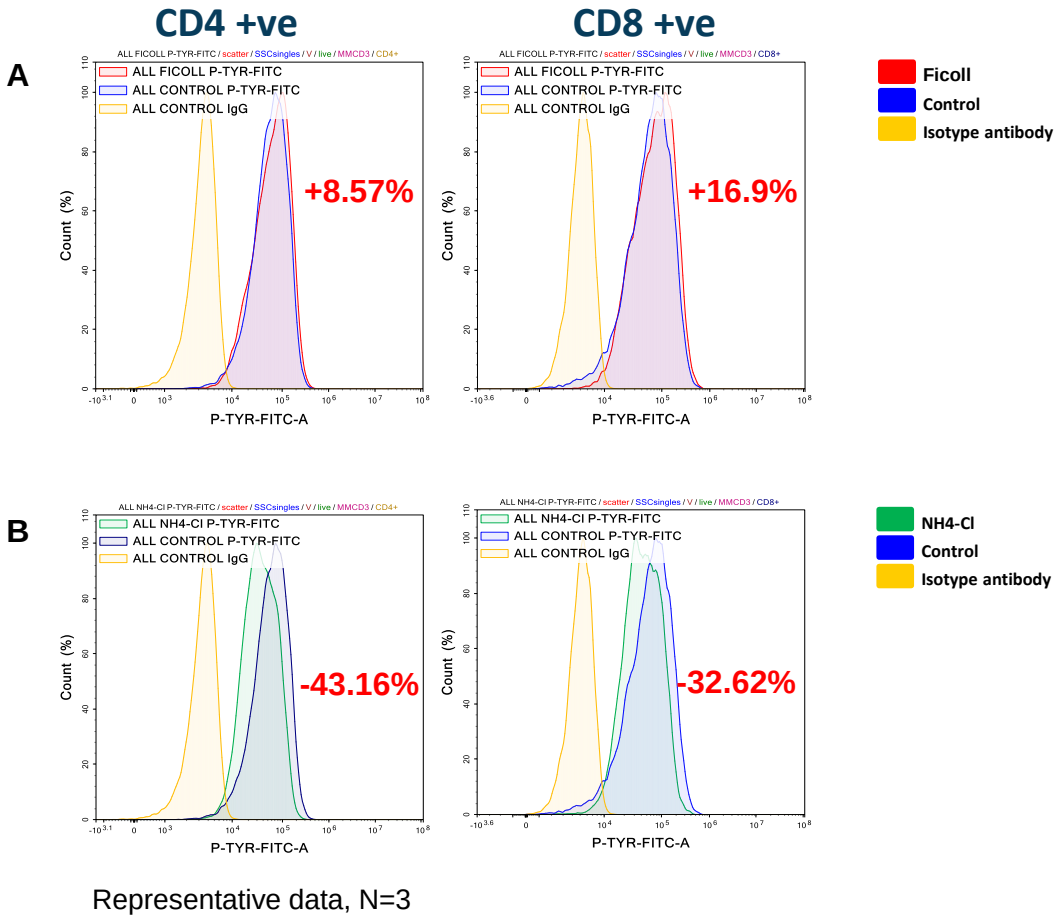
Non-DCS T cells have significantly lower mass vs. DCS processed, then negatively selected T cells

Ratio of Mitobrilliant staining normalized to DCS preparations and compared over time.

DCS prepared T cells show a less altered phosphorylation profile vs. Ficoll or Ammonium Chloride Exposed Cell

Fitness for Purpose

- Pathway Biology
- Avoidance of Chemicals
- Avoidance of cell-cell contact



Representative data, N=3

- DCS-prepared PBMC were thawed and exposed to cell separation reagents
- Differential phosphorylation was evaluated immediately after

A. Ficoll Protocol results in ~8-17% difference in shift[1]

B. 10' Ammonium Chloride exposure followed by washing impairs 30-40% of T cell phosphorylation[2]

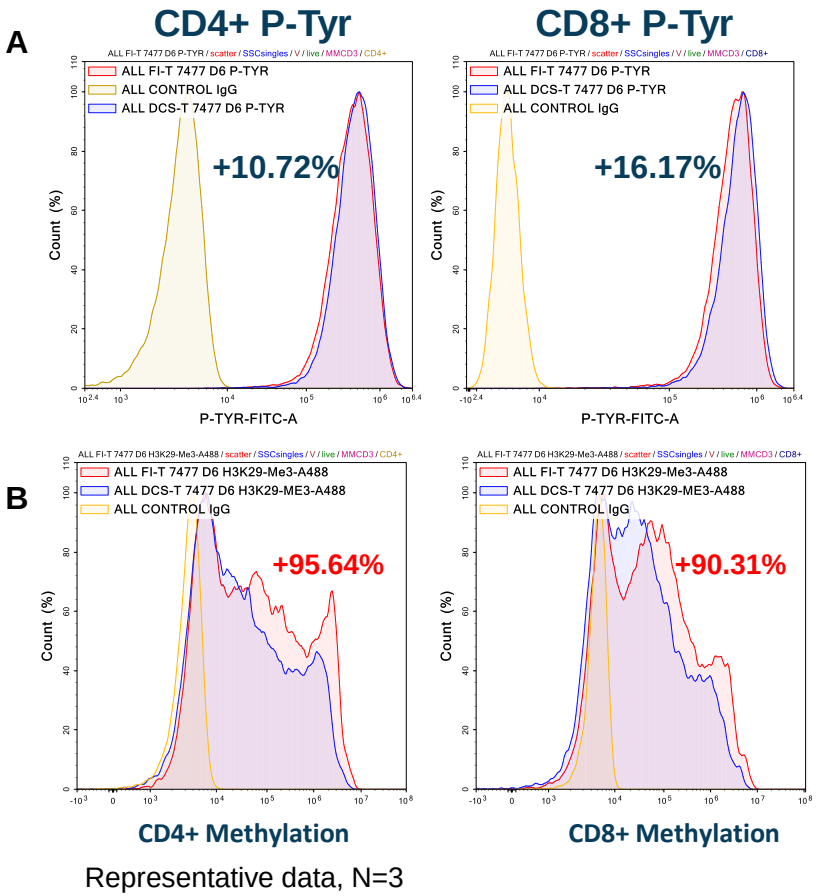
Exposure to Separation Agents impacts cell signaling appreciably

1. This observation is largely consistent with Curate's internal data showing that Ficoll treated cells have increased activation vs. DCS prepared cell
2. Response to oxidative stress

Post thaw processing confirms that PBMC are epigenetically redirected

Fitness for Purpose

- Pathway Biology
- Avoidance of Chemicals
- Avoidance of cell-cell contact



Representative data, N=3

DCS cells were frozen in CS-10, thawed after 1 Mo and left unseparated or Ficolled and then expanded (Transact, IL-7/-15).

A. % phosphorylation indicates that DCS cells are slightly more responsive.

B. Thawed DCS PBMC T cells less methylated[3] than Ficoll prepared PBMC

Differential methylation persists at Day 6 post thaw/expansion

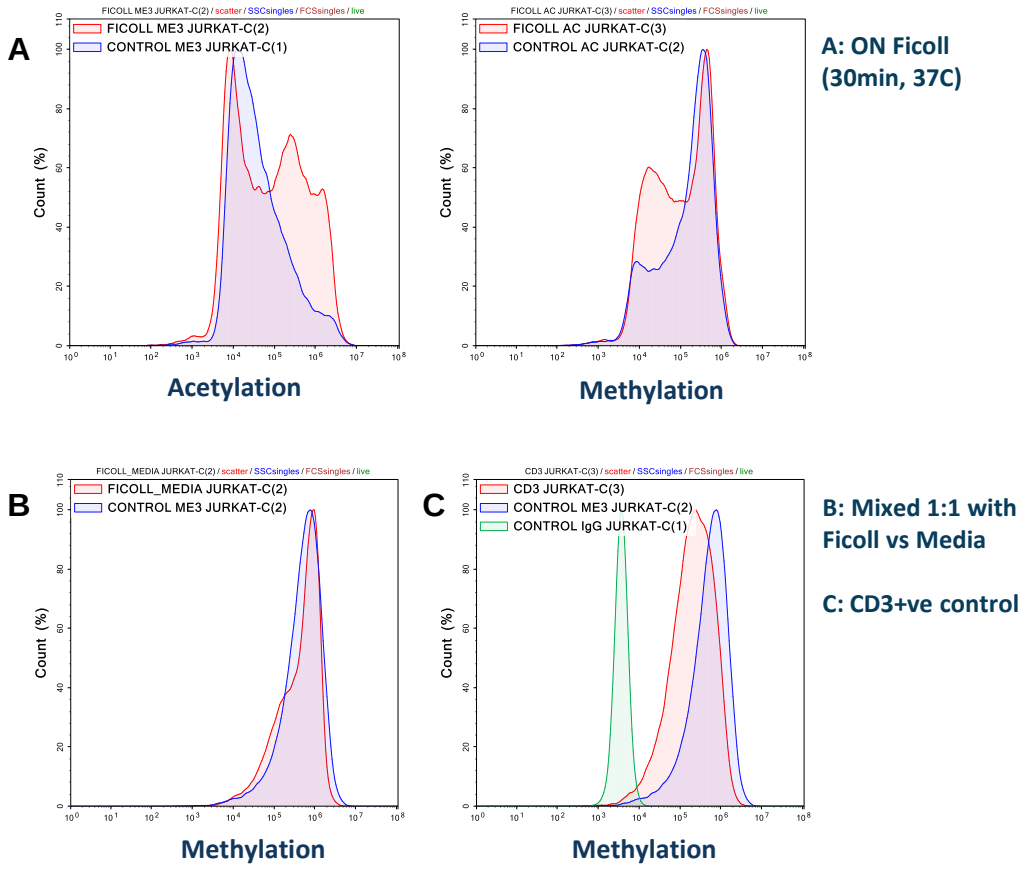
Affirms epigenetic change

3. Araki et al. Genome-wide Analysis of Histone Methylation Reveals Chromatin State-Based Regulation of Gene Transcription and Function of Memory CD8+ T Cells. Immunity 30, 912–925, June 19, 2009 DOI 10.1016/j.immuni.2009.05.006

Cell:Cell contact during centrifugal processing is a **significant** driver of methylation state

- **Fitness for Purpose**
  - Pathway Biology
  - Avoidance of Chemicals
  - Avoidance of cell-cell contact

T cell line (Jurkat) Trimethylation of Histone3 Lysine27 by PRC2



A: ON Ficoll (30min, 37C)

B: Mixed 1:1 with Ficoll vs Media

C: CD3+ve control

Representative data, N=3

Hypomethylation of Histone 3 Trimethylation in K27 position is associated with open chromatin position and "ON" for transcription

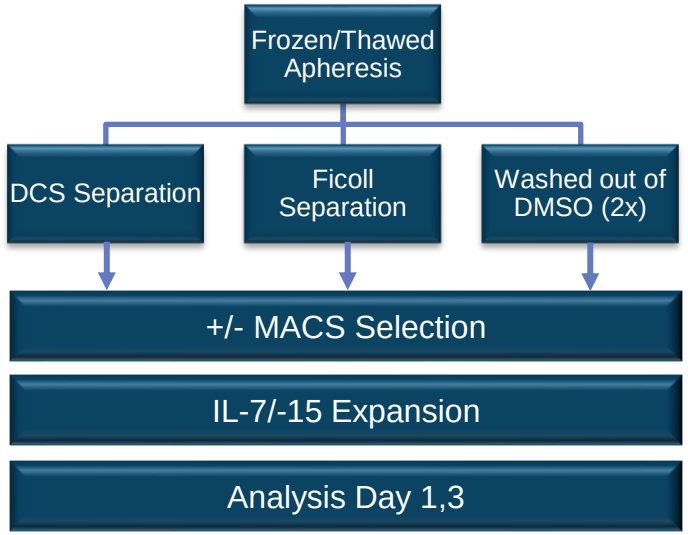
A. **Ficoll exposure to Jurkat cells results in significant methylation/acetylation shift**

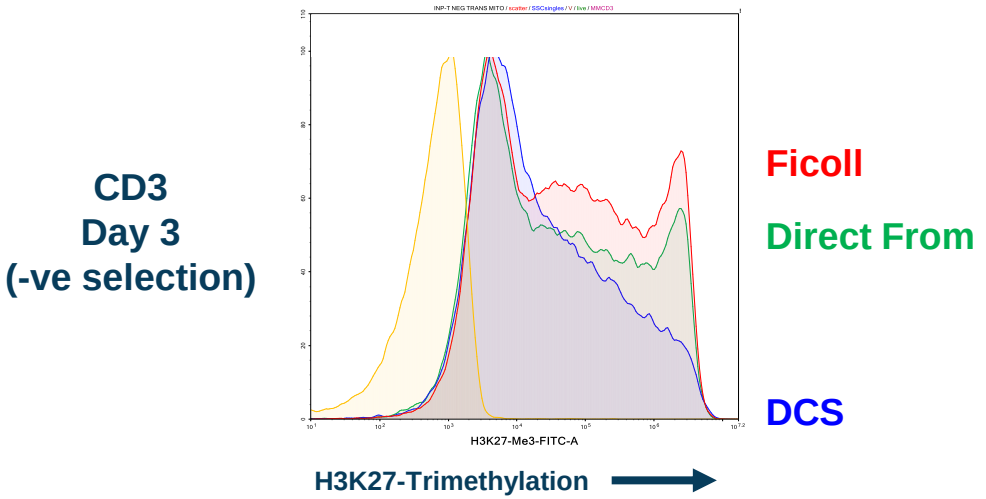
B. **Jurkat cells mixed 1:1 with Ficoll (no gradient) induces little/no change in methylation**

C: Effects of Antibody Binding (+ve control)

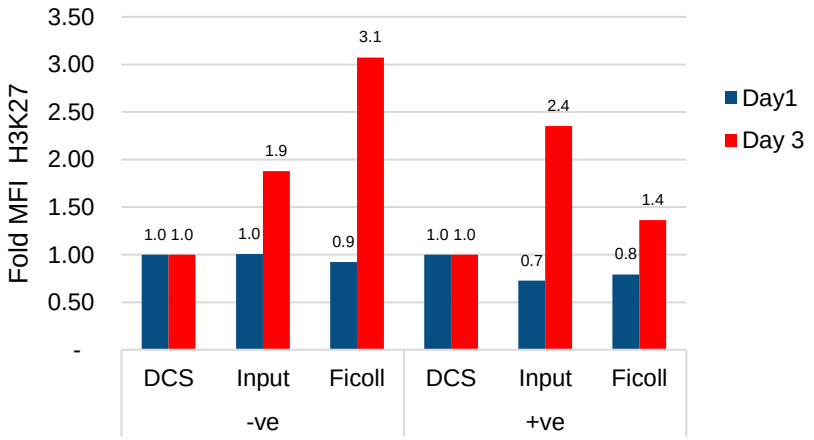
Increased cell-cell interaction at Ficoll interface is a significant driver of histone modification

Processing and Environment Drive Differential Methylation in isolated T cells

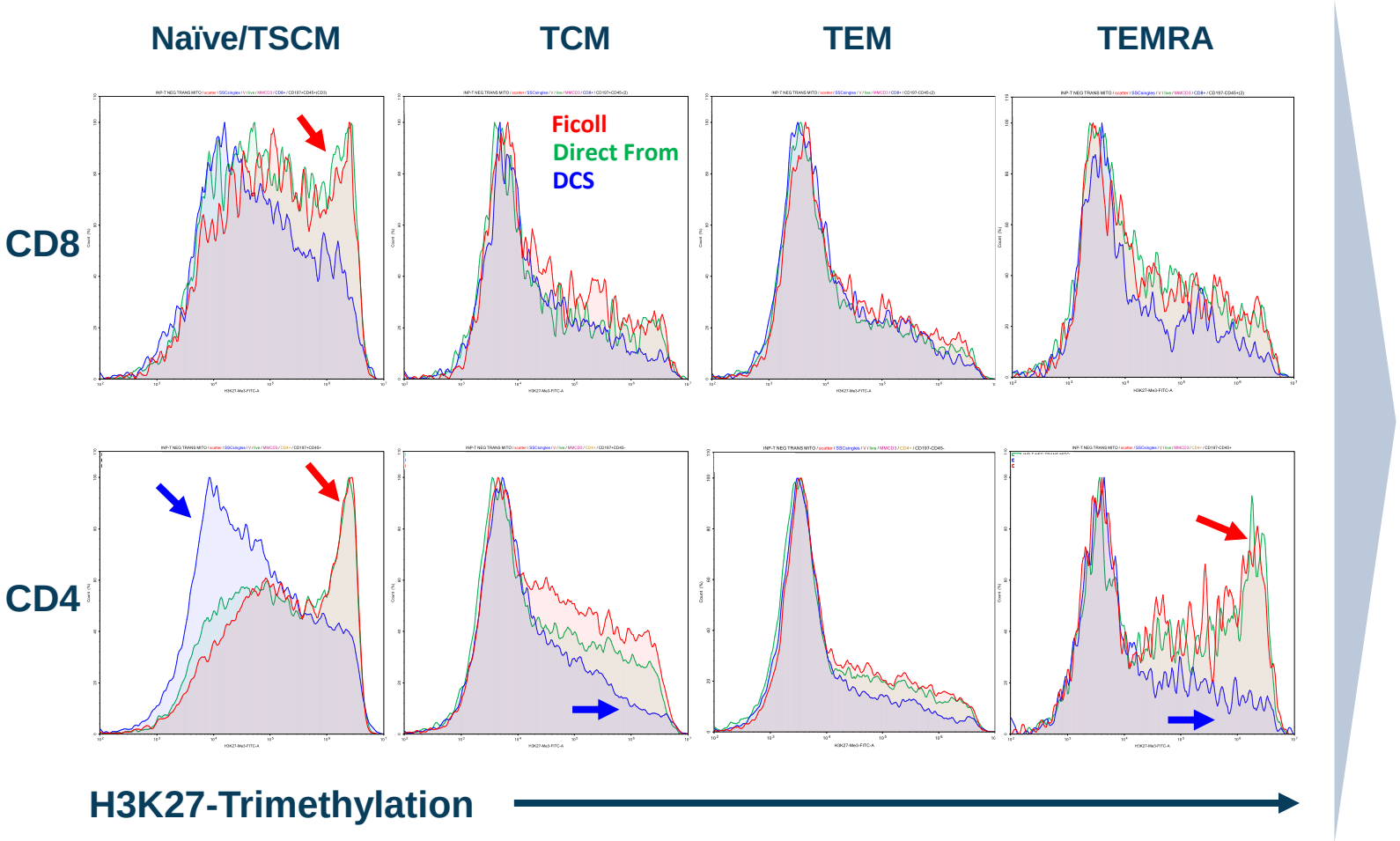




- Observation
  - Purified T-cell methylation shifts as a function of sample preparation methodology
  - Positive and Negative Selection impact methylation states differently (likely due to antibody binding effect)



Environment & processing drive Histone (H3K27) tri-methylation changes in T cell memory subtypes

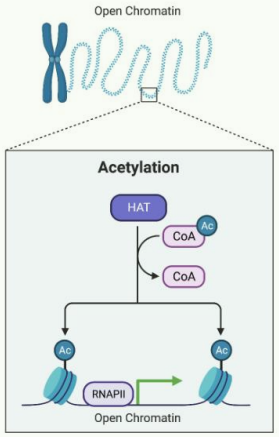


- Observation
  - Hypo-methylation bias particularly present in CD4, CD8 naïve T cells
  - CD4+ Tcm , Temra also significantly lower
- DCS cells are uniquely less methylated vs. **both** Ficoll/Direct Selection
  - Consistent with platelet/cytokine removal

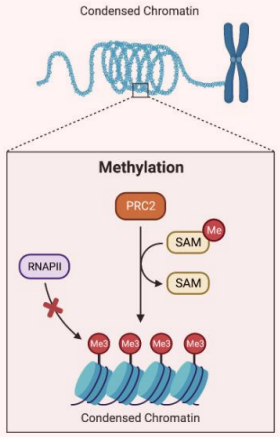
Histone Methylation is a highly conserved potent regulator of Gene Silencing, Cell Differentiation

DCS

Centrifuge

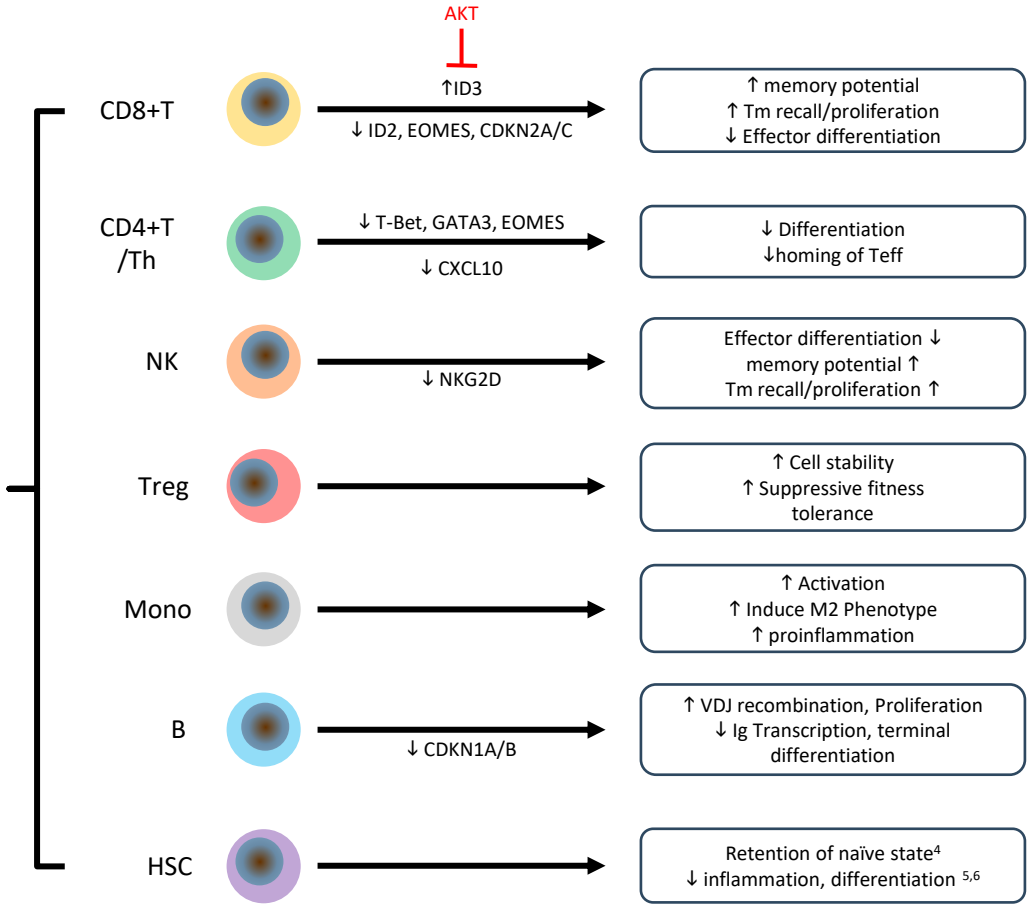


Transcription "ON"

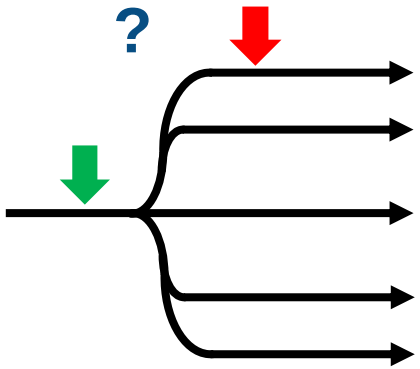


Transcription "OFF"

H3K27Me3



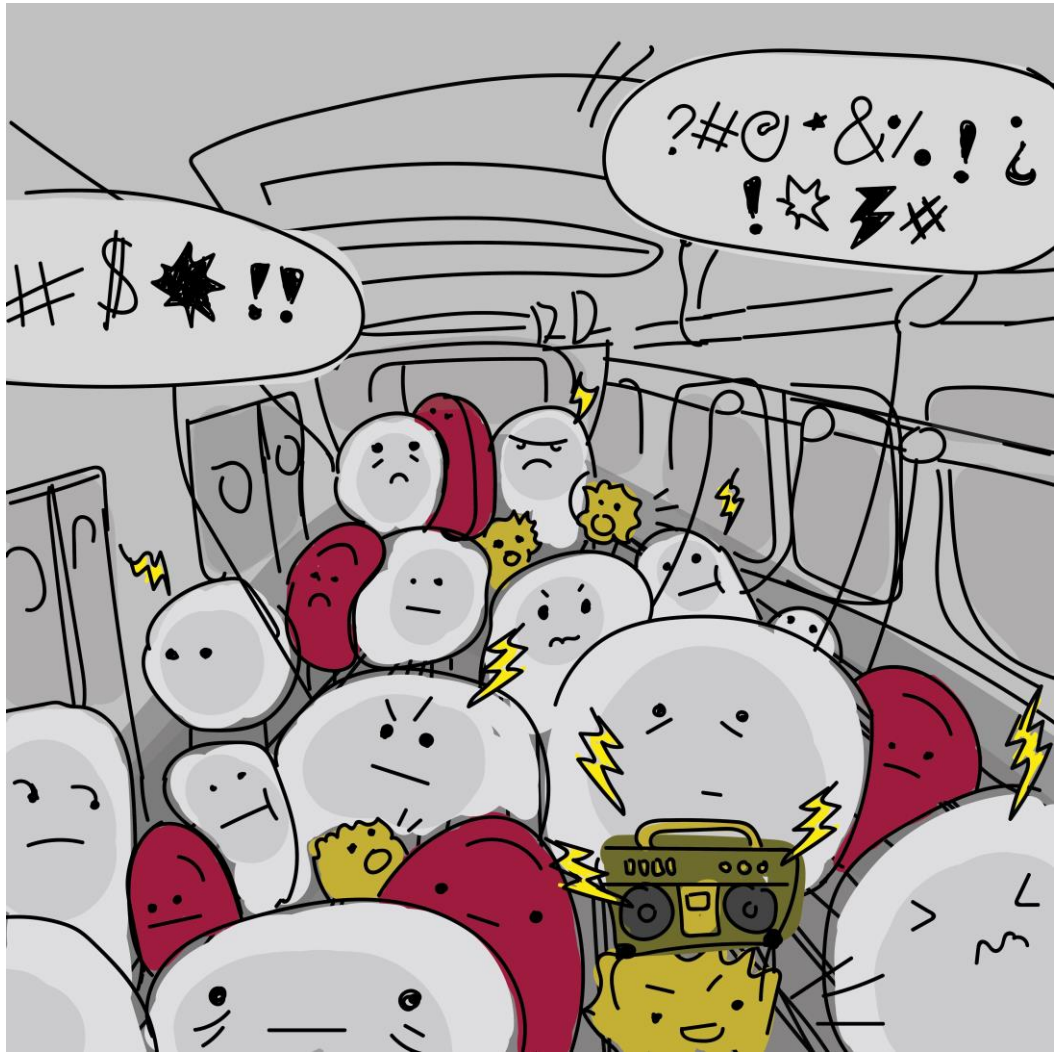
Potential impact: ALL Significant CGT cell Targets

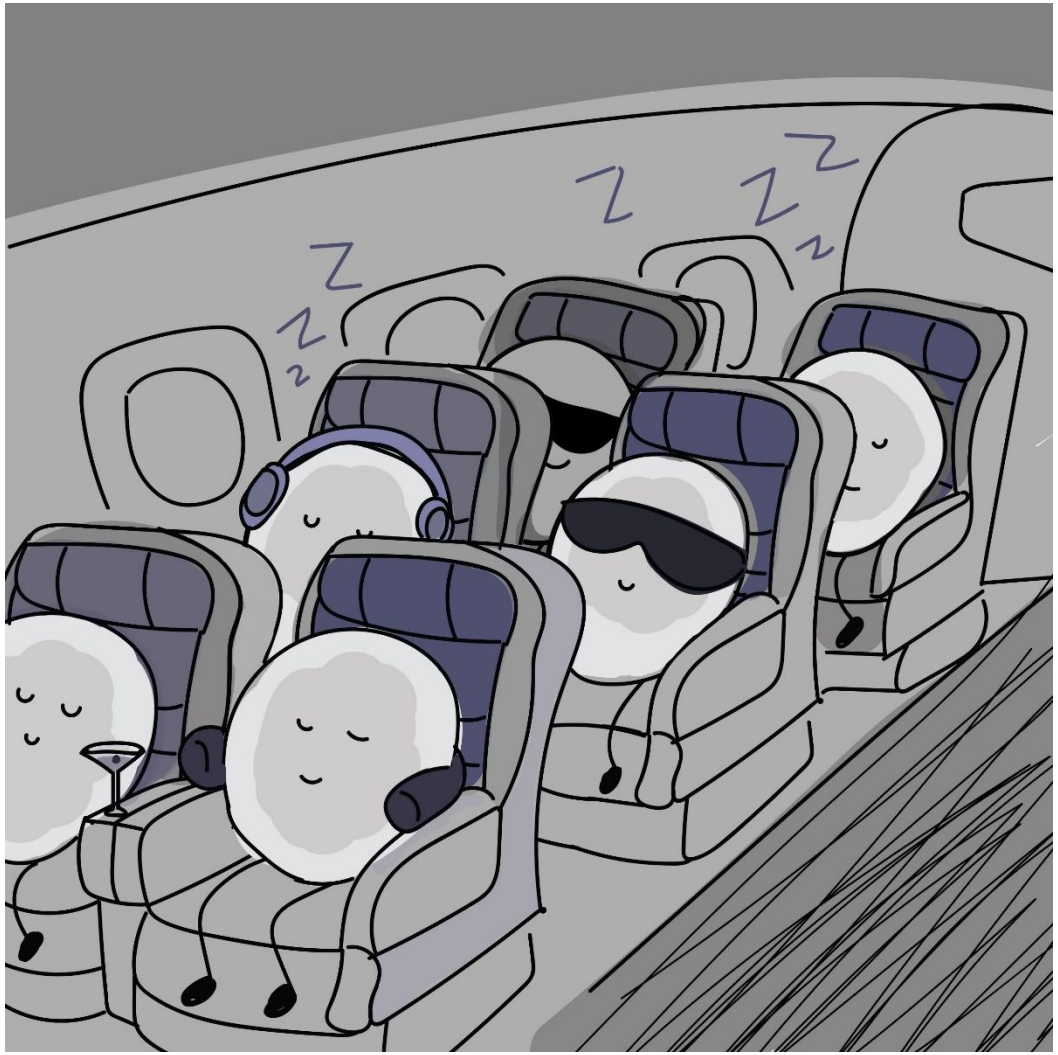


Figures Adapted from Biorender, The functions of EZH2 in immune cells: Principles for novel immunotherapies, J Leukocyte Bio. 10.1002/JLB.1RU0520-311R

4. Long, Y. *et al.* RNA is essential for PRC2 chromatin occupancy and function in human pluripotent stem cells. *Nat Genet* **52**, 931–938 (2020)
5. Kfoury-Beaumont, N. *et al.* The H3K27M mutation alters stem cell growth, epigenetic regulation, and differentiation potential. *BMC Biol* **20**, 124 (2022)
6. Schuettpelz, L et al. Regulation of hematopoietic stem cell activity by inflammation Front. Immunol., 19 July 2013 https://doi.org/10.3389/fimmu.2013.00204

How do you want your cells to arrive when you start processing?





Curate DCS cells are metabolically and epigenetically superior vs. centrifuged cells

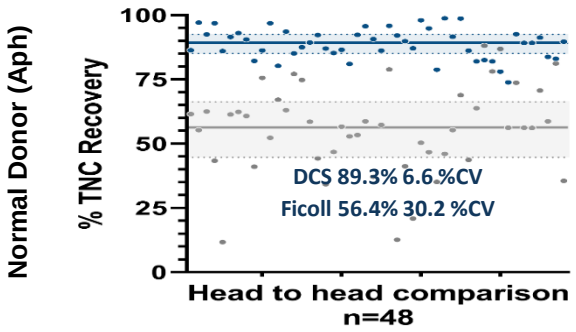
- DCS cells are uniquely fit for purpose
- All data aligns with higher clinical efficacy
  - Highest metabolic potential
  - Most retention of Naïve Tscm/Tcm phenotype
    - Less generation of Treg
  - Physically and more epigenetically available Naïve CD4+ cells are available for transcription

- Metabolically more fit
  - Up to 40% more mitochondrial mass
- More able to signal properly
  - No Oxidative stress response
    - Up to 40% more phosphorylation vs Ammonium Chloride exposed cells
  - Less activation
    - Up to 15% less phosphorylation vs Ficoll processed cells
- Lower DNA Methylation state
  - DCS cells likely **start** in a similar state as retraining with Dasatinib/Tazemetostat which target PRC2/EZH2/H3K27 Trimethylation
    - CAR-T "reversed" to Tscm status with drug pulses improves efficacy and potency in clinical studies, but takes days/weeks to achieve[1]
  - Hypomethylation phenotype seen in DCS vs. all centrifugal approaches tested

1. Weber et al. Transient rest restores functionality in exhausted CAR-T cells through epigenetic remodeling. Science . 2021 Apr 2;372(6537):eaba1786. doi: 10.1126/science.aba1786.

Curate's technical advantage is derived from three discrete areas:

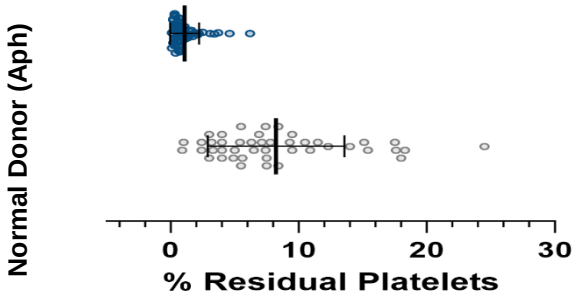
Physical Recovery



~2x Unbiased Recovery

No bias/cell loss

Cell Separation Quality



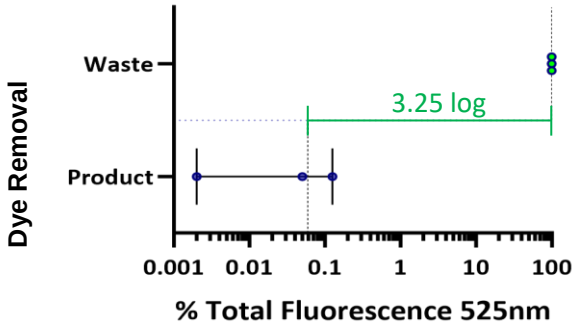
>14x fewer platelets

>3 fold fewer RBC

No chemicals

No colocalization

3 Log Wash in <1 sec



Rapid and complete non centrifugal removal of Cytokine, Chemokines, etc.

The right cell is available …

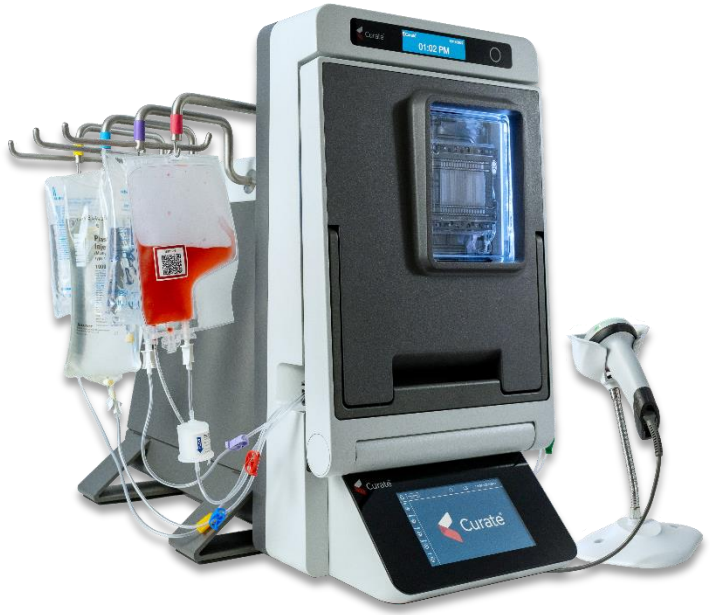
… is free from:

- forced cell-cell contact
- epigenetically active agents
- cells that generate biology and process impairment

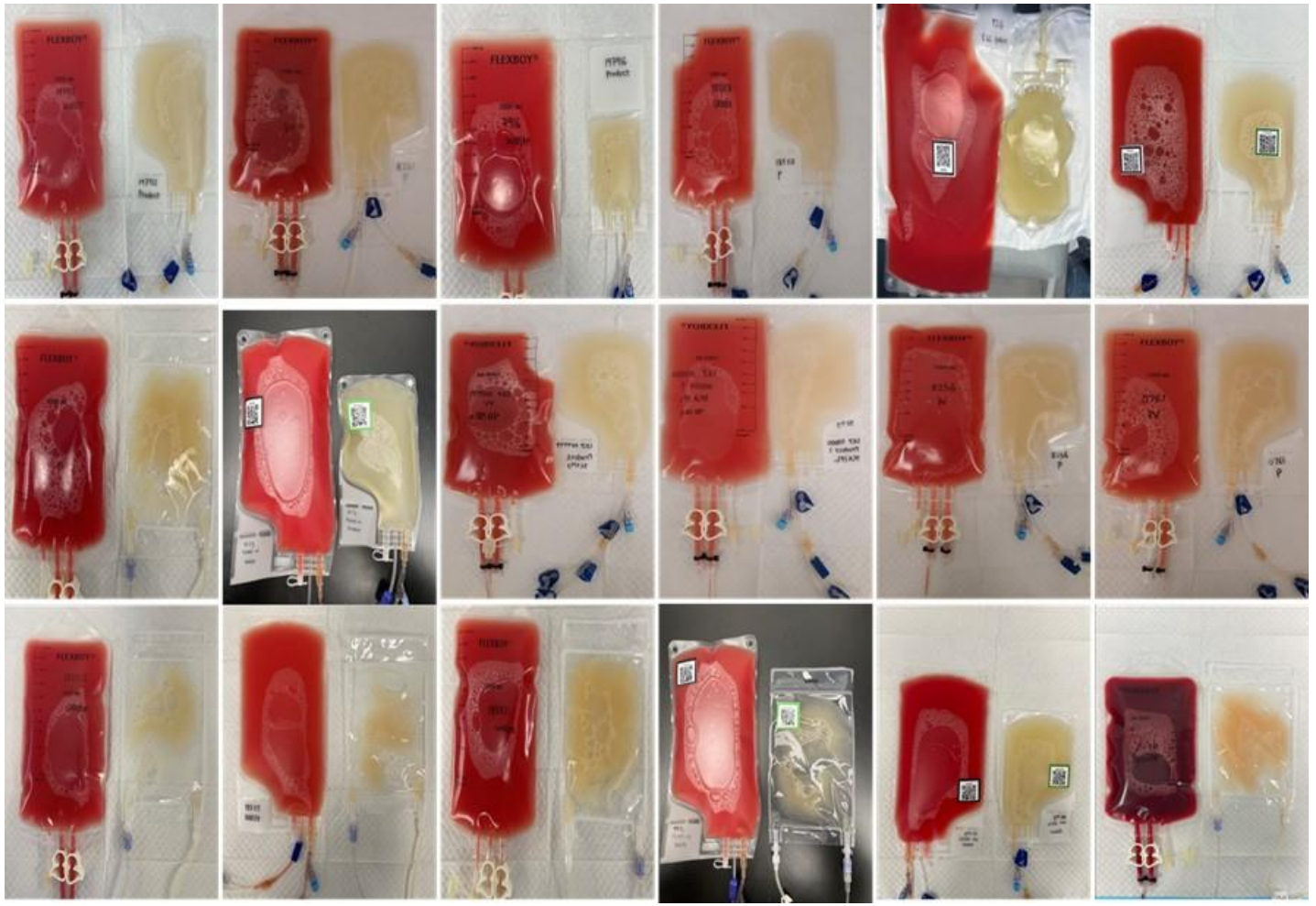
… is free from soluble factors that:

- Impair virus
- Predetermine differentiation

The Curate System delivers uniquely well-prepared cells with uniquely good consistency…



- Functionally closed
- Easy to use, minimal hands-on time
- Processes **unfiltered** leukopak to PBMC in <1 hour
- cGMP-grade solution
- 21-CFR-11, Networkable



techsupport@curatebio.com