Implementation of the Cue for a Cell Therapy Manufacturing Process

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CTMC + a joint venture between Resilience + MD Anderson Cancer Center

Introduction

CTMC is a joint venture between National Resilience and MD Anderson. Through their collaborative partnership model, CTMC provides process development and integrated regulatory support with the aim of bringing novel cell therapies seamlessly and efficiently through every step of the development journey.

The final formulation, fill, and finish unit operation in the manufacturing of cell and gene therapies is an essential part of the process. Current methods typically require manual processing, which carries the risk of operator error and variability. The Cue Cell Processing System from Fresenius-Kabi provides an automated solution for this processing step. This system has the capabilities to concentrate, wash, dilute, formulate, and aliquot cell suspensions. The single use consumable allows for functionally closed processing, suitable for GMP manufacturing. A device master file (DMF) has been filed with the FDA to facilitate use of the system in a clinical and/or commercial process.

Figure 1: The Cue Cell Processing Instrument

Table 1: System Key Features

Instrument Features	
Separation Technology	Spinning membrane filtration, 4um pore size
Volume Accuracy	±10% or 1 mL, whichever is greater
Solution Types	Up to 2 wash buffers & 1 formulation buffer Verified solution density range: 1.01 – 1.125 g/mL
Formulation Strategy	100% resuspension into formulation buffer; Ability to add solutions in a configurable ratio (e.g. 1:1)
Maximum Number of Aliquots	100
Minimum Final Volume	10mL for bulk volume 2mL minimum aliquot volume
Aliquot Bag Type	Configurable

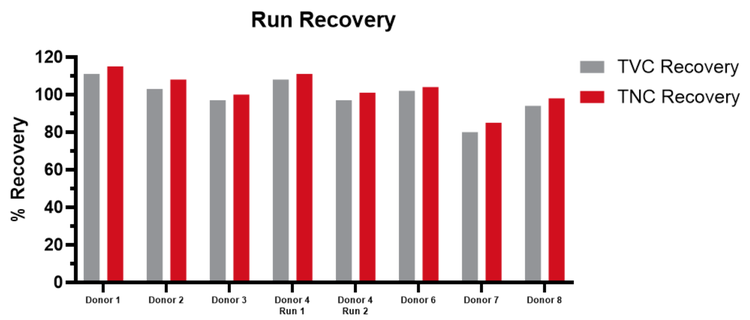
Because of Cue's capabilities, CTMC designed experimentation to verify and validate the system for use in manufacturing processes that require the concentration, wash, dilution, formulation, and aliquoting of culture expanded T-Cells to prepare for cryopreservation.

Methods

- T-Cells were isolated, engineered, and subsequently expanded from 9 donors (not all healthy, patient material was used for Donor 3). From these 9 donors, 11 Cue procedural runs were completed.
- On harvest day, cells were harvested from the bioreactor into a transfer pack that was sterile welded to the Cue Cell Processing System primary kit.
- The cells were processed using a custom designed protocol for CTMC that was designed to wash out spent culture media, resuspend cells into Plasmalyte+5%HSA, and add CS10 in a 1:1 ratio to reach a defined viable CAR-T+ concentration.
- Volume was measured pre procedure using a weigh scale. The Cue reported value was recorded as the volume at the post-formulation state (after the addition of CS10).
- Post formulation, the protocol was designed to fill cryo- bags. Cell counts and viability samples were measured pre and post Cue procedure using the NC-200.
- The viability was also measured for the post thawed aliquots for all runs except Donor 6.

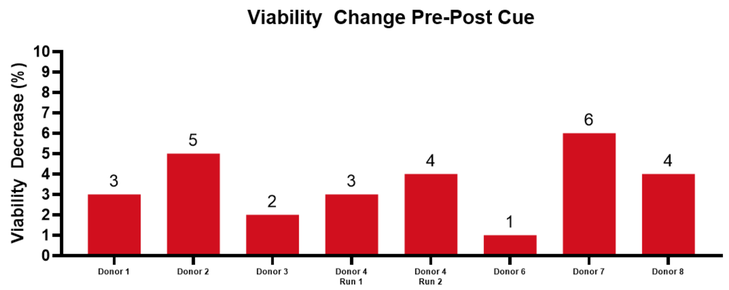
Procedural Results

Plot 1: TNC and TVC Recovery Pre-Procedure to Post Cue Formulation



- Plot 1 shows the TNC and TVC recoveries, which were calculated using the measured NC-200 concentrations of the pre-procedure and post Cue formulation samples.
- Recoveries were calculated for 8 out of the 11 total runs.
- Some runs observed >100% recovery, which could be due to clumps of cells that are uncounted in the pre-procedure sample that are counted in the post-procedure sample. Spinning membrane filtration, which is the core separation technology of the Cue, breaks apart cell clumps. Additionally, the Cue reported volume is subject to the volume accuracy of the system.

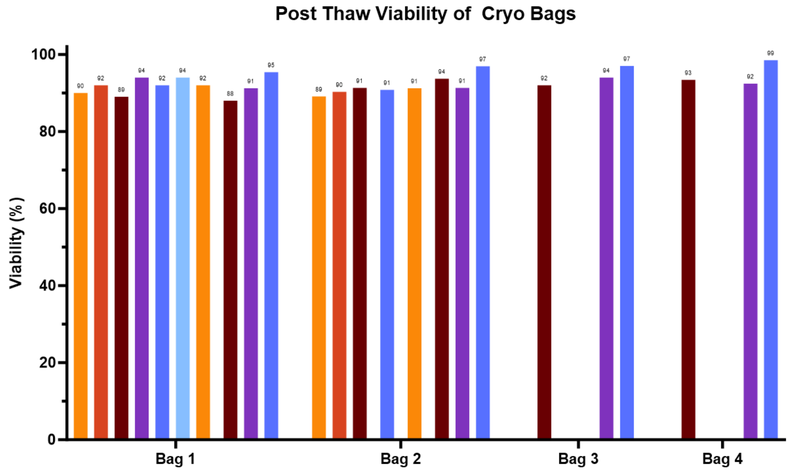
Plot 2: Viability Change from Pre-Procedure to Post Cue Formulation



- Plot 2 shows the viability change of the cell suspension, pre-procedure to post formulation state. The average viability drop across all runs was 3±0.0164%.

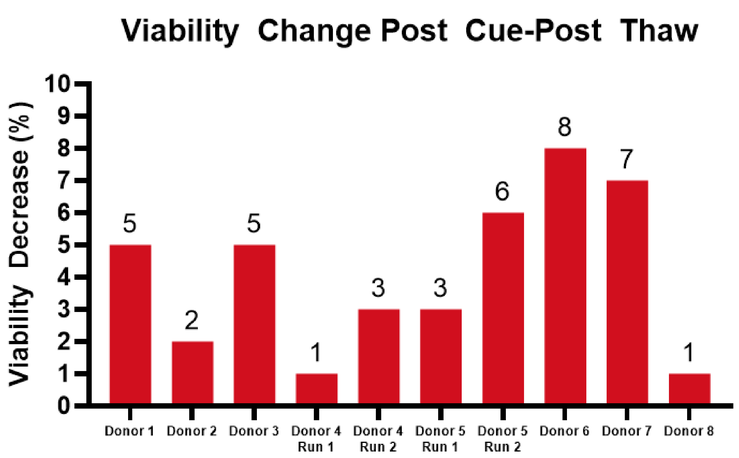
Post Thaw Results

Plot 3: Post Thaw Viability



- Plot 3 displays the measured viability of the cryo-bags post thaw.
- Some runs had multiple bags that were filled and frozen.
- Viability was measured immediately post-thaw.
- Overall, the average post thaw viability was 92.47±0.0254% across all cryo-bags and runs.

Plot 4: Viability Change from Post Cue Procedure to Post Thaw



- The average drop in viability from the end of the Cue procedure to post-thaw was 4±0.0229%.

Conclusions

- The average TVC recovery calculated across runs for the Cue procedure was 98.93±0.0966%.
- All cell counts and viability measurements met the defined requirements of this unit operation.
- The Cue Cell Processing System from Fresenius-Kabi provides an automated, functionally closed solution for cryopreservation preparation of engineered T-cells for streamlined integration into a clinical or commercial cell therapy manufacturing process.