A Novel Acoustic Cell Processing Platform For CAR-T Cell Concentration

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A Novel Acoustic Cell Processing Platform

1. Acoustic standing wave is generated within flow chamber. The standing wave creates a multi-dimensional acoustic field.

2. Lateral acoustic forces create tight clusters of particles, which continuously grow, as more particles are concentrated in the cluster.

3. Flow Direction

4. When particle clusters reach a critical volume, enhanced gravitational settling causes particle clusters to settle out into a collection port.

A NOVEL ACOUTIC CELL PROCESSING PLATFORM

Flow Direction

Flow Direction

Flow Direction

Flow Direction

No sonication/cavitation

No sonication/cavitation

No sonication/cavitation

No sonication/cavitation

PROCESS OVERVIEW AND SPECIFICATION

Existing pain points for the concentrate-wash step for cell therapy include low cell recovery, long wash cycles with large buffer volumes and open-process manual manipulation and complexity, i.e., difficult to scale up or out as well as automate for commercial manufacturing.

PROCESS OVERVIEW

1. Cultured cells from bioreactor vessel or bag are pumped into acoustic element.
2. Cells are captured and concentrated in the acoustic element. Clarified media is discarded.
3. Wash fluid or new media is pumped into the acoustic element.
4. Old media is pushed out of the acoustic element and cells are re-suspended in wash fluid.
5. Concentrated cells are recovered from element and collected for further processing.

PROCESS SPECIFICATIONS

- Feed Volumes 7 500 mL to 3L
- Processing Time 7 60 Minutes
- Incoming Feed Density 7 < 1M cells to 40M cells per mL
- Target Viable Cell Density Recovery (VCD) 7 80%
- Effect on Cell Viability 7 None
- Final Concentrate Volume 7 7 ml @ 1M/mL, >50ml @ 40M/mL
- Cell Concentration Factor 7 15X @ 40M/mL to 140X @ 1M/mL
- Processing Temperature Rise 7 < 7.5°C

FUNCTIONALLY CLOSED CONSUMABLE KIT

- Acoustic Element
  - Materials: Polycarbonate and Stainless Steel
  - Single-use design
- Tubing Kit
  - 1/8" PVC thin-wall tubing
  - Able to sterlize weld feed bags to the kit for cell processing
- Current kits use a single-use pulseless pump head
  - NanoStedi 2.5x5ml - disposable dosing pulseless pump head
- Kit & Element are Double-Bagged, Gamma Irradiated
- Process Flow Diagram
  - Allows for priming, recirculation, concentration, media exchange or washing and collection
- Current System Limitations
  - Sized for feeds of up to 3L
  - Total cell capacity 7 4 - 8 billion cells
  - Final concentrated volume 7 6 - 50 mL
- V: Solenoid Valve
  - R: Air-Liquid Sensor
  - T: Temperature Sensor

RESULTS - ACOUTIC CONCENTRATE-WASH (ACW)

- Table 1 shows the input, output and performance experimental values for a low surface density (1LE-1, 1.5 E6/mL) ACW volume reduction. Figure 1 depicts the viable cell density (VCD) and viability of the primary cultures of T-cells after ACW processing.
- Table 2 displays the input, output and performance experimental values for a high surface density (1LE-1, 1.5 E6/mL) ACW volume reduction. Figure 2 shows the effect of power on Viable Cell Recovery (VCR) using the high cell density 1LE-1 element at a flow rate of 30mL/min.

PRODUCT ROADMAP

- Development system for early stage customer trials and process optimization
- Q1 2018: Co-development systems to be placed on-site for concentrate wash trials
- Q4 2018: Flexible commercial system for multiple unit operations, scales, and process parameters

CONCLUSIONS & FUTURE WORK

- The acoustic cell processing platform offers a robust cell concentration and washing unit operation for cell therapy applications.
- QC analytical results have shown that cells are not impacted by exposure to the acoustic process.
- Development of the technology to handle full spectrum of feed volumes, cell types and cell concentrations.
- Define, characterize and optimize technology and parameters towards a more robust cell processing platform for use in affinity cell selection.

VCR of High Cell Density Runs 1LE vs. Power

Figure 1 – Viable cell density (VCD) and viability of primary cultures of T-cells after ACW processing. The cells from each 1LE experiment were re-seeded at 1E6/mL, 37°C, 5% CO₂ in duplicate and counted 24h later.

Figure 2 – The effect of power on Viable Cell Recovery (VCR) using the high cell density 1LE-1 element at a flow rate of 30mL/min.