Application Note

Scalability: Consistent and reproducible manufacturing across a multi-IRO® system.

Overview

IRO® is a next generation platform that automates and standardizes critical steps in cell and gene therapy manufacturing. Its core innovations include a bellows-based Bioreactor with customizable operation modes (static, rocking and compression mixing), and OriConnect™ tubeless sterile connection for automated fluid handling.

This technical note summarizes data demonstrating IRO's ability to deliver consistent results when the starting material and process inputs are kept the same. Data from five donors confirm reproducibility across multiple IRO platforms, based on key metrics including T cell expansion, transduction efficiency and phenotype.

Summary

- T cells from the same donor were manufactured in parallel across multiple IRO platforms and cell growth and quality attributes (transduction efficiency and phenotype) were compared.
- IRO demonstrated high reproducibility across multiple platforms, with low inter-platform variability (coefficient of variation [CV]) including viable cell yield (<3%), transduction efficiency (<5%) and phenotype (3-7%).

Assessing manufacturing reproducibility across multiple IRO platforms

Isolated CD3+ T cells from five different donors were manufactured in parallel across multiple IRO platforms using a standardized process, each starting with 150 million T cells from the same bulk preparation. Cell count and viability were monitored daily from day 5 until harvest. Transduction efficiency and T cell phenotype were analyzed using flow cytometry. Inter-instrument variability was assessed by calculating the coefficient of variation (CV) across cell quality attributes.

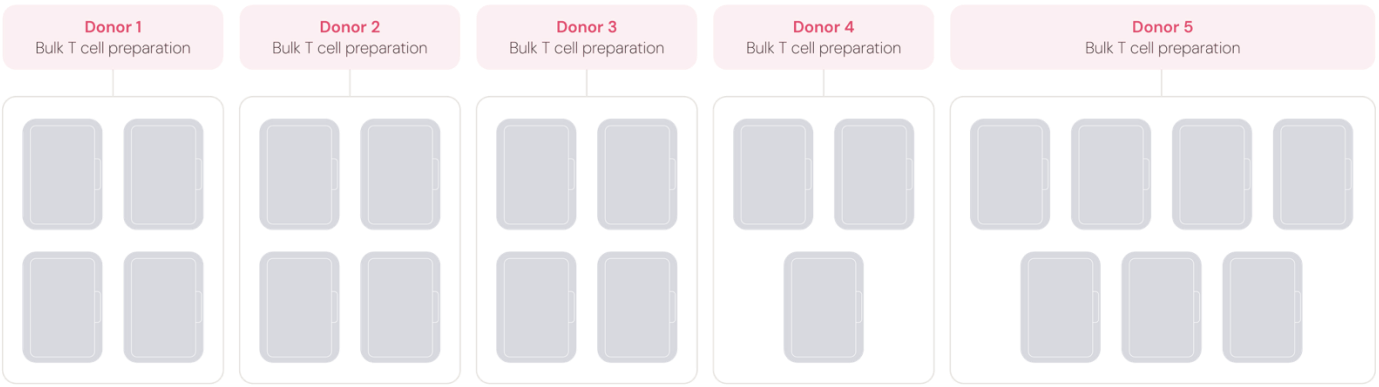

Figure 1. Testing the reproducibility of T cell manufacturing across multiple IRO platforms. T cells were isolated from 5 donors. For each donor a bulk cell preparation was made and then T cells were split into parallel IRO platforms for manufacturing (3–7 IROs per donor, as shown above). Cell growth, transduction efficiency and phenotype of T cell products from the same material split across multiple IRO platforms was compared (see Figure 2 and Table 1).

Results

T cell manufacturing in IRO demonstrated consistent performance across multiple platforms. For each donor, total viable cell yield was highly consistent with a CV between instruments of <3% across multiple process days (Figure 2). In addition, reproducibility was demonstrated across instruments for all five donors when looking at transduction efficiency, CD4+ and CD8+ T cell frequencies and T cell memory phenotype (Figure 2).

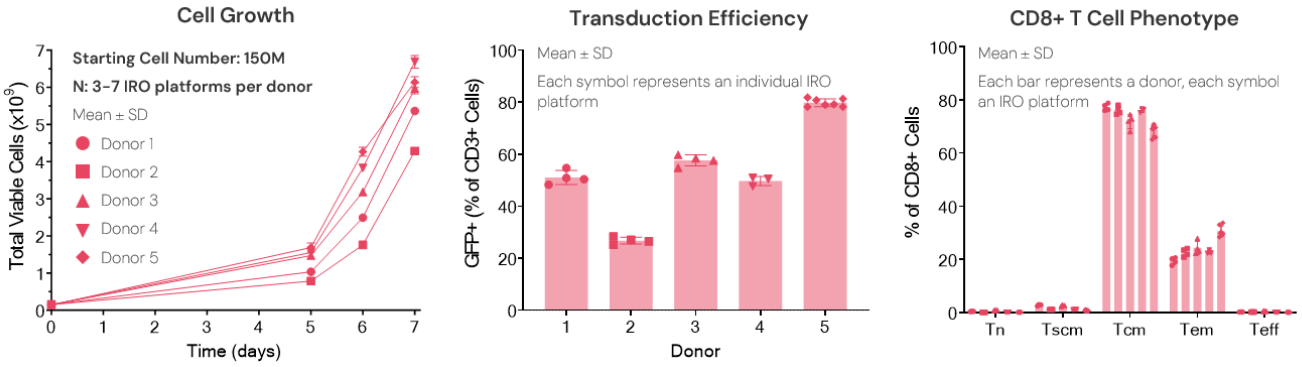

Figure 2. Reproducible T cell manufacturing across multiple IRO platforms. Cell growth, transduction efficiency and T cell phenotype from T cell products manufactured from the same T cell bulk material split across multiple IRO platforms were compared. Shown above is the cell growth (mean total viable cells across all IRO platforms ± standard deviation [SD]), transduction efficiency (mean GFP+ [% of CD3+ cells] ± SD) and T cell phenotype (mean [% of CD8+ cells] ± SD). Tn [naïve], CD45RO−CCR7+CD95−; Tscm [stem cell memory], CD45RO−CCR7+CD95+; Tcm [central memory], CD45RO+CCR7+; Tem [effector memory], CD45RO+CCR7−; Teff [effector], CD45RO−CCR7−.

Healthy Donor #	# of IRO platforms tested	Average data (coefficient of variation across platforms)				
		TVC x10^9	Transduction efficiency (%)	CD4+ (% of CD3+)	CD8+ (% of CD3+)	Tscm + Tcm (% of CD8+)
Donor 1	4	5.3 (0.8)	50.6 (3.6)	59.9 (2.8)	38.8 (4.2)	80.4 (1.6)
Donor 2	4	4.7 (1.5)	26.8 (4.9)	71.7 (5.1)	20.9 (3.3)	77.2 (2.0)
Donor 3	4	4.3 (2.4)	57.7 (3.7)	40.3 (6.9)	52.1 (2.0)	75.2 (3.5)
Donor 4	3	3.1 (2.6)	49.7 (3.5)	64.6 (1.7)	34.1 (3.1)	77.2 (0.9)
Donor 5	7	6.0 (2.4)	79.8 (1.9)	74.8 (0.5)	23.3 (1.5)	69.4 (2.9)
All donors	**22**	**4.7 (max 2.6)**	**52.9 (max 4.9)**	**62.3 (max 6.9)**	**33.8 (max 4.2)**	**75.9 (max 2.9)**

Table 1. Summary of inter-platform variation in IRO. T cells from five donors were manufactured in parallel in 3 to 7 IRO platforms. Key metrics on the day of harvest were compared, including cell growth (total viable cells [TVC]), transduction efficiency and T cell phenotype. In the table above we show average data for TVC, transduction efficiency, CD4+ (% of CD3+), CD8+ (% of CD3+) and Tscm/cm (% of CD8+). The coefficient of variation (CV) for each of these analytics is captured in the parentheses. All metrics show less than 7% CV across platforms. Tscm, T stem cell memory, CD45RO-CCR7+CD95+; Tcm, T central memory, CD45RO+CCR7+.

Conclusions

- IRO reliably supports scalable T cell manufacturing with consistent yields across multiple IRO platforms, resulting in a coefficient of variation <3% for viable cell counts at all sampling points.
- Key metrics like viable cell yield, transduction efficiency, and T cell phenotype are highly reproducible across platforms.

With highly reproducible and consistent biological performance IRO delivers the benefits of automation to maximize clinical and commercial impact.

Scale Your Impact

To learn more and access additional resources, please visit oribiotech.com/iro or scan the QR code

About Us: Ori Biotech is a London and Philadelphia-based manufacturing technology company pioneering flexible process discovery with seamless translation and scalable commercialization of cell and gene therapies.

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