A Rapid Approach to Streamlined CART Therapies and Reduced Cost of Goods

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Abstract

Introduction

As cell therapy becomes more prevalent, reducing cost of goods (COGs) and increasing manufacturing batch success becomes more pressing. Finding mechanisms to facilitate this reduction in COGs and manufacturing failures is critical to improving access to life saving cell therapy treatments and the continued uptake of these transformative therapies. Kincell Bio recently partnered with Syenex to evaluate their RapidCell™ engineered vector to rapidly manufacture CD19-targeting CART cells. Syenex RapidCell™ is an advanced gene delivery vector engineered to selectively target and activate specific cell types while efficiently mediating transduction. The RapidCell™ vector for T-cell engineering eliminates the need for external activation reagents and cell isolation steps traditionally required in CART cell manufacturing, resulting in a streamlined production workflow.

Methods

Using an industry standard cell therapy platform, Kincell Bio evaluated the Syenex RapidCell™ engineered vector at multiple MOIs across multiple donor leukopaks, which were platelet washed prior to transduction. Cell mass was then harvested and cryopreserved prior to further analysis.

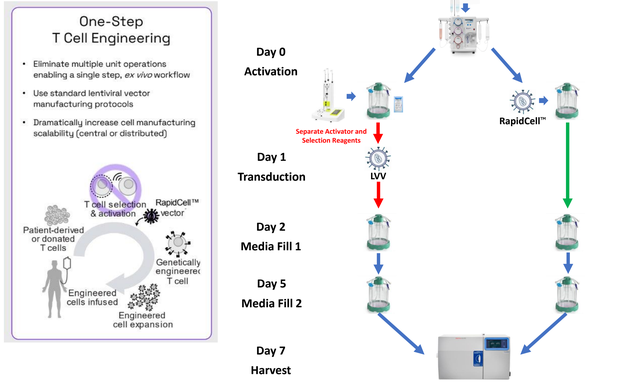
Results

We found that the RapidCell™ engineered vector integrated smoothly into existing Kincell Bio process development unit operations, while also reducing processing time by approximately 40%, including the removal of entire operations. This resulted in a ~55% reduction in estimated cost of goods based on use of the RapidCell™ engineered vector. These savings, in labor and raw materials occurred in the context of robust cell growth, high CAR positivity, high T-cell purity, and minimal differentiation.

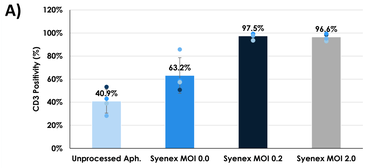
Conclusion

As cell therapy products, including CART products, continue to demonstrate efficacy in a growing range of indications, new critical raw materials will need to evolve to meet demand, cost, and access requirements. Syenex RapidCell™ represents a significant step toward closing the gap in access to advanced therapies. Here, we demonstrate the novel use of Syenex RapidCell™ technology to manufacture a CD19 targeting CART product that displayed the desirable attributes of high purity, CAR expression, expansion, and potency.

Graphical Abstract

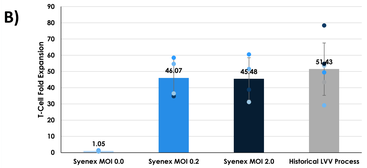


RapidCell™ Based T-Cell Activation Leads to Robust Cell Growth and High T-Cell Purity



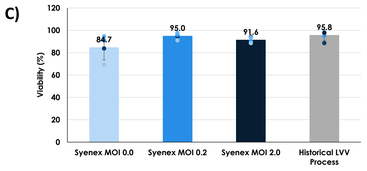
A. Syenex RapidCell™ vector selectively activates CD3⁺ T-Cells, ultimately leading to high CD3⁺ T-Cell Purity at harvest

- CD3⁺ purity greater then 95% in all cases when Syenex RapidCell™ vector used
- Comparable purity at MOI 0.2 and 2.0



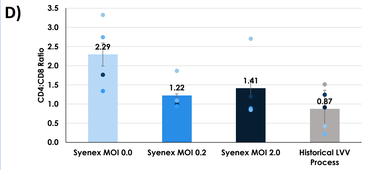
B. Syenex RapidCell™ vector CD3⁺ T-Cell activation induces expansion comparable to the historic LVV process

- On average the RapidCell™ process is within 10% of the historic LVV Process
- Comparable expansion at MOI 0.2 and 2.0

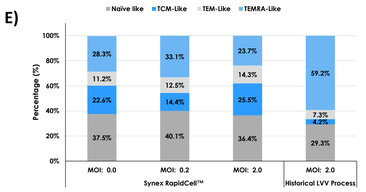


C. Viability at harvest consistently higher then 88% when cells were activated with Syenex RapidCell™ vector

- No significant differences observed among activated conditions



D. No significant difference in CD4:CD8 ratio observed between the Syenex RapidCell™ vector and the historical LVV based process

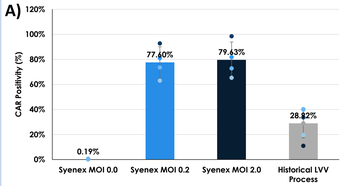


E. Syenex RapidCell™ vector activation effectively conserved starting T-cell subset distribution

- Comparable T-cell subset distribution at MOI 0.2 and 2.0
- At both MOI values tested, the Syenex RapidCell™ vector had a smaller population of more exhausted TEMRA-like cells then the historical LVV process

- Each point represents 1 donor (N of 4 for Syenex conditions, N of 6 for historical LVV process). Bar represents the average of donors tested. Error bars represents one standard deviation.

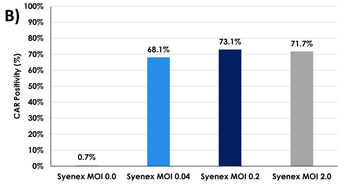
RapidCell™ Vector Generates a Potent CART Product with High CAR Positivity



A. Syenex RapidCell™ vector increased transduction percentage by at least 2.6-fold over the historical LVV process

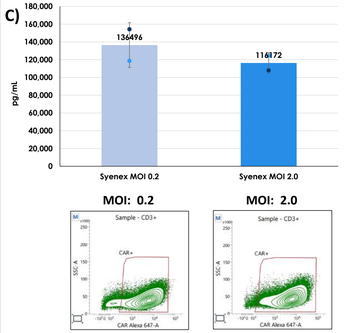
- Average CAR positivity of 77.6 and 79.63% using Syenex RapidCell™ vector
- Comparable CAR⁺ percentage at MOI 0.2 and 2.0

- Each point represents 1 donor (N of 4 for Syenex conditions, N of 6 for historical LVV process). Bar represents the average of donors tested. Error bars represents one standard deviation



B. High CAR⁺ in the 0.02 MOI condition prompted a follow up study testing a lower MOI, in this case 0.04 (N of 1)

- It was found that even at this lower MOI CAR positivity was 68.1%, a decrease of only 5%
- This data suggests that at an MOI of 0.2 the system may be near saturation and that even lower MOIs may be efficacious

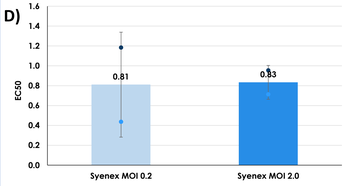


C. Potency was assessed by measuring IFN-γ release in Syenex process–produced samples

- Antigen-specific IFN-γ secretion reached ~10^5 pg/mL, with ≤3% background in control conditions with measurements within the assay linear range

- To support complex analytics two representative samples were tested for each condition. CAR positivity of the respective samples are below, example Donor A CAR plots shown

CAR Positivity		
MOI	Donor A	Donor B
Syenex 0.2	92.77%	81.08%
Syenex 2.0	98.45%	81.85%



D. As an additional assessment of potency, a cell killing assay was performed on the same two conditions

- EC50 values ranged from 0.45 to 1.2 (E:T) across all samples, within the range commonly observed for CART products

- To support complex analytics two representative samples were tested for each condition. CAR positivity of the respective samples are below, example Donor A CAR plots shown

Syenex RapidCell™ Technology is Purpose-Built to Enable Lower CART Costs

A. Leveraging the ability of Syenex RapidCell™ vector to activate and transduce T-Cells, batch cost can be reduced by approximately 50%

- Syenex RapidCell™ vector eliminates the need for cell selection and a separate transduction operation, leading to an approximate 40% reduction in manufacturing operations
- Largely driven by the lack of selection and of a separate activation reagent, an estimated material cost savings of 55% is expected

	Syenex Process
Labor Cost	-40%
COGS	-55%
Cost per Batch	**-50%**

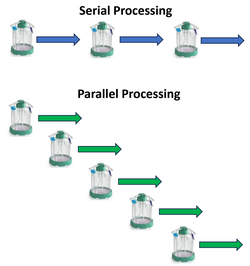
B. Syenex RapidCell™ vector further reduces cost per dose when CAR⁺% is considered

- Based on a 2.0E+08 CAR⁺ VC dose (2.5E+06 CAR⁺ VC/kg, 80 kg patient), the Syenex RapidCell™ vector produces 11 doses to the 4 of the historic process
- An approximate 82% decrease in cost per dose

	Historic Process	Syenex Process
Doses per Batch	4	11
Cost per Dose		**-82%**

C. Parallel processing is expected to magnify the potential of the Syenex RapidCell™ vector to drive down cost by increasing throughput and optimizing resource utilization

- Asset and GMP suite utilization: Parallel batch processing minimizes equipment down time and maximizes cleanroom throughput
- Labor productivity: Standardized, parallel operations enable multi-batch oversight, reducing labor cost per dose
- Throughput efficiency: Parallelization reduces scheduling gaps and increases batch cadence, driving greater output per unit time with potential for less then proportional cost increase
  - Additional potential to automate as relevant technologies mature



Discussion

Kincell Bio has demonstrated the efficacy of Syenex RapidCell™ vector and established the capacity of Syenex RapidCell™ vector to integrate easily into existing CART manufacturing paradigms. Kincell Bio assessment of Syenex RapidCell™ vector suggests the following benefits from its usage:

- Cost per batch decrease of ~50% and a per dose reduction of ~80%
- Decreased vector material needs per batch, reducing the need for additional vector lots and associated lot bridging activities
- Improved or comparable product characteristics to the historical LVV process
  - CD4:CD8 ratio, CD3 positivity, CAR positivity, T-Cell subset distribution
- Industry aligned product potency as measured by two-methods

Syenex RapidCell™ vector has the potential to substantially reduce CART manufacturing costs and operational burden. As CART therapies become more widely adopted, innovations like Syenex RapidCell™ vector will be essential to ensuring patient access to CART therapies.

Acknowledgments

We would like to acknowledge the Kincell Bio Process Development team (Isabella Mejia-Ruiz, Prarthana Ravishankar, and Rachel Nosacka-Boucher) for operational support and thoughtful discussion and the Kincell Bio Analytical Development team (Dominique Aponte, Jessica Wehinger, Gennarino Del Bagno, Danielle Kling, Patrick Kellish) for analytical support.