Abstract #1163 | ISCT 2026

Synecta™ T1 Cell-Derived Nanoparticles Enhance Lentiviral Transduction and T Cell Manufacturing Efficiency with IRO®: A Closed, Automated Cell Therapy Manufacturing Platform

Chiquita Hanindya[1] · William Raimes[2] · Jessica Weaver[2] · Peter Keller[1]

[1]BlueWhale Bio, Inc., Philadelphia, PA · [2]Oribiotech Ltd, London, UK

KEY FINDINGS

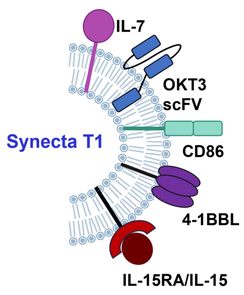
1. Synecta T1 + IRO achieved the top transduction and yield performance amongst comparators.
2. A 48-hour static hold in IRO maximized Day 7 CAR-T cell output.
3. Performance was reproduced across independent runs, supporting platform and workflow robustness.
4. Synecta T1 shifted T cells toward a fitter, less glycolysis-skewed metabolic state.

ABSTRACT & BACKGROUND

CAR-T cell therapy manufacturing remains constrained by two interconnected bottlenecks:

1) Activation strategies that impair cell fitness, metabolic state, and transduction readiness.
2) Static culture systems that limit expansion and gene delivery.

Conventional CD3/CD28 bead-based workflows add complexity through removal steps, ongoing cytokine/media supplementation, and delayed transduction, creating challenges for both cell quality and scalable manufacturing. **Synecta T1** cell-derived nanoparticles (CDNP) are a biodegradable, bead-free activation platform designed to improve activation biology through localized, multi-signal stimulation. By eliminating bead removal, reducing reliance on exogenous cytokines, and enabling Day 0 activation and transduction, **Synecta T1** supports a favorable activation state, improved T-cell fitness, and a metabolic profile associated with productive expansion. Complementing this biology-driven approach the **IRO®** platform provides a closed, automated manufacturing environment with programmable fed-batch culture, dynamic mixing modes and real-time process insights to support improved transduction and expansion relative to fully static workflows. **Together, Synecta T1 and IRO® address both biological and process bottlenecks in CAR-T manufacturing, pairing a transduction-supportive activation strategy with an automation-enabled system for scalable production.**



Schematic of Synecta T1 cell-derived nanoparticles (CDNPs), a biodegradable, feeder-and-bead-free activation platform displaying membrane-bound OKT3 scFv, CD86, 4-1BBL, IL-7, and IL-15/IL-15Rα to deliver localized, multi-signal T-cell activation.

IRO, the new standard in CGT manufacturing, automates and accelerates cell therapy manufacturing, increasing throughput, improving quality and decreasing costs by combining proprietary hardware, consumables, software, data and analytics.

METHODS

Condition	Growth Vessel	Reagent	Protocol Applied
C1	IRO Platform	Synecta T1	24h Static Culture (only) Post-Seed
C2			48h Static Culture (only) Post-Seed
C3			Rock Mix Post-Seed (No Static Period)
C4		Comparator 2	24h Static Culture (only) Post-Seed
C5	Static Bioreactor	Synecta T1	Static Culture Throughout
C6		Comparator 2	Static Culture Throughout

IRO Bellows Bioreactor

Synecta T1 Conditions

- Transduced on Day 0 regardless of the growth vessel.
- No soluble Cytokines added to the growth medium.

Comparator 2 Conditions

- Comparator 2 (Comp.) is a widely used Polymeric Nanomatrix reagent.
- Transduced on Day 1 regardless of the growth vessel.
- Cytokines added : 5ng/mL IL-7 + 10ng/mL IL-15.

Day 0: Seed and Activate

- Thaw and Seed CD3+ T cells: 150M into IRO and 15M into Static Bioreactor
- Add Synecta T1 and Lentiviral Vector (LVV) to C1, C2, C3 and C5
- Add Comparator 1 to C4 and C6

Days 0-2

- Start Mixing IRO Bioreactor on:
  - Day 0 in C3 (i.e. post-seed)
  - Day 1 in C1 and C4
  - Day 2 in C2

Day 1

- Add LVV to Comparator 1 cultures C4 and C6

Days 3-6

- Automated IRO Sample and Feeds:
  - Daily sample for viable cell count
  - Sequential feeds to reach 1000mL
  - Transition from Rock to Compression

Day 3

- Manual Sample of Static Bioreactor for cell count, then Media top-up to max working volume 100mL

DAY 7: Harvest and Analytics

- Harvest final products from IRO and Static Bioreactor.
- Final sample for cell count and total viable cell yield.
- Conduct Flow Analysis on final product: Transduction efficiency (GFP+ or CD19+), CD4/CD8 ratio and memory phenotype (CD45RA, CCR7, CD95).

RESULTS

Figure 1 — Synecta T1 Outperforms in Total GFP Transduced Cell Yield Across Both Bioreactors with Highest Yield in IRO

Figure 1A: Fold Expansion

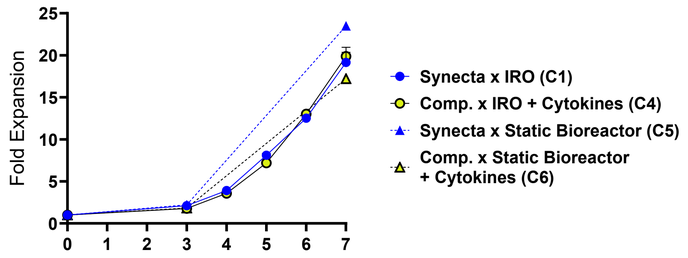


Fig. 1A. T cell fold expansion over time in IRO and static bioreactor culture formats for Synecta T1 and Comparator 2 across dynamic IRO and static bioreactor conditions. N = 2 for IRO conditions and N = 1 for static bioreactor conditions.

Figure 1B: Transduction Efficiency on Day 7

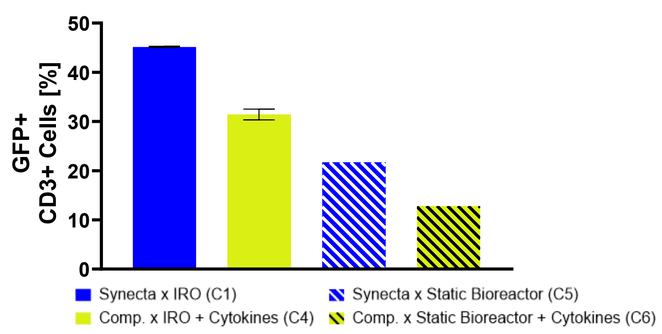


Fig. 1B. Day 7 Transduction efficiency of T cells cultured in IRO and static bioreactor formats measured for Synecta T1 and Comparator 2. MOI = 1; N = 2 for IRO conditions and N = 1 for static bioreactor conditions.

Figure 1C: Total Viable GFP+ Cells on Day 7

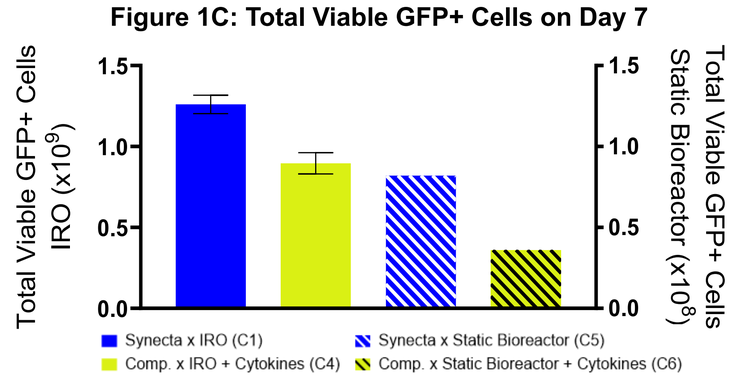


Fig. 1C. Day 7 Total viable GFP+ T cell yield in IRO and static bioreactor measured for Synecta T1 and Comparator 2. MOI = 1; N = 2 for IRO conditions and N = 1 for static bioreactor conditions.

Figure 1D: CD4/CD8 Population on Day 7

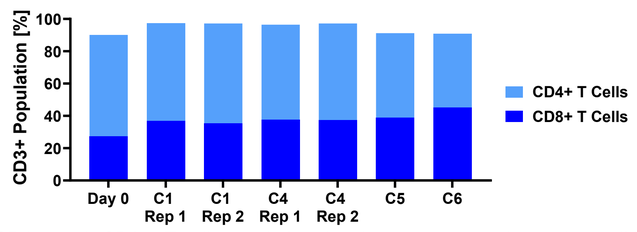


Fig. 1D. CD4+ and CD8+ T-cell population levels across experimental conditions were assessed on Day 0 and Day 7 across tested samples. N = 2 for IRO conditions and N = 1 for static bioreactor conditions.

Figure 1E: T Cell Memory Phenotype on Day 7

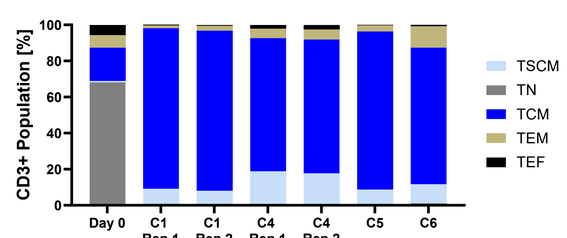


Fig. 1E. Memory T-cell subset distribution based on CD45RA, CCR7, and CD95 expression on Day 0 and Day 7 across experimental conditions were evaluated for the indicated IRO and static culture conditions. T_N, naïve T cells; T_{SCM}, stem cell memory T cells; T_{CM}, central memory T cells; T_{EM}, effector memory T cells; T_{EF}, effector T cells. N = 2 for IRO conditions and N = 1 for static culture conditions.

T cell expansion was robust in both IRO and static culture formats, with Synecta T1 supporting greater expansion than Comparator 2 in the static bioreactor. This advantage became most evident in transduction performance and total viable transduced cell output, where **the combination of Synecta T1 and IRO produced the highest overall T-cell yield across all conditions tested**. The improved yield reflects the benefit of pairing Synecta T1-mediated activation with the dynamic, automated culture environment of the IRO system. Importantly, this stronger manufacturing performance was achieved without major changes in end-of-culture phenotype, as **CD4/CD8 composition remained balanced** and **T cell memory phenotype subsets were broadly comparable across conditions**. Together, these findings demonstrate that Synecta T1 is highly compatible with the IRO platform without any workflow optimization. **Synecta T1 combined with the IRO bioreactor supports the strongest overall cell yield while maintaining key phenotypic features of the final T cell product.**

Figure 2 — Extended Static Activation (C2: 48h Static) Achieves the Highest CAR-T Cell Yield in IRO

Figure 2A: Fold Expansion

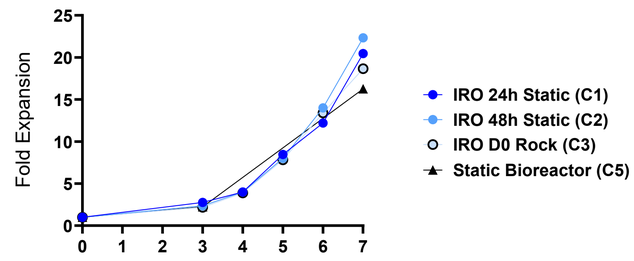


Fig. 2A. Total viable cell count over time across IRO mixing conditions and static culture were tracked at the indicated time points for C1 (IRO 24 h static), C2 (IRO 48 h static), C3 (IRO Day 0 rock), and C5 (static culture). N=1.

Figure 2B: Total Viable CD19+ Cells on Day 7

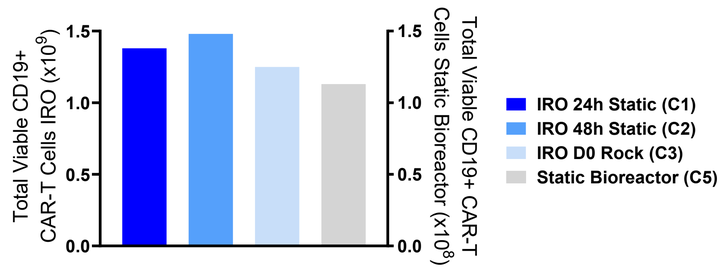


Fig. 2B. Total viable CD19+ CAR-T cell count on Day 7 across IRO mixing initiation conditions and static culture. Total viable CD19+ CAR-T cells were measured on Day 7 for C1 (IRO 24 h static), C2 (IRO 48 h static), C3 (IRO Day 0 rock), and C5 (static bioreactor). MOI = 0.5; N=1.

Figure 2C: CD4/CD8 Population on Day 7

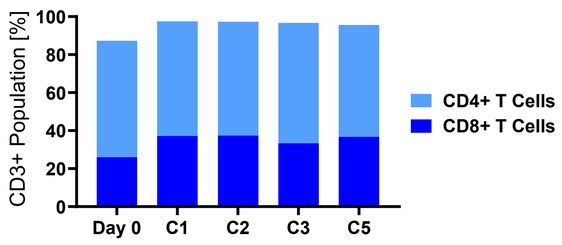


Fig. 2C. CD4+/CD8+ T-cell population distribution on Day 7 across IRO® mixing initiation conditions and static culture. CD4+ and CD8+ T cell population levels were assessed on Day 0 and Day 7 for C1, C2, C3, and C5.

Figure 2D: T Cell Memory Phenotype on Day 7

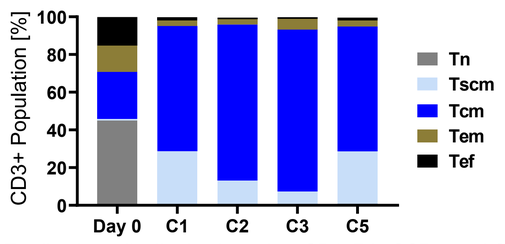


Fig. 2D. Memory T cell subset distribution based on CD45RA, CCR7, and CD95 expression on Day 7 across IRO mixing conditions and static culture. Memory phenotype profiles were measured for Day 0, C1, C2, C3, and C5. T_n, naïve T cells; T_{scm}, stem cell memory T cells; T_{cm}, central memory T cells; T_{em}, effector memory T cells; T_{ef}, effector T cells.

Varying the timing of transduction mixing within the IRO workflow influenced final manufacturing output, with the 48-hour static condition (C2) producing the highest total viable cell expansion and the greatest total viable CD19+ CAR-T cell count at harvest. The 24-hour static condition (C1) also performed strongly, while Day 0 rocking (C3) showed lower yield than the early-static IRO conditions. **Notably, the IRO bioreactor conditions yielded higher overall CD19+ CAR-T cell output than the static bioreactor (C5) highlighting the advantage of the closed, automated system.** CD4/CD8 composition remained broadly balanced across conditions, and memory phenotype profiles were preserved, with central memory T cells as the predominant subset at Day 7.

Figure 3 — Synecta T1 Activation Shifts T Cell Metabolism Toward Oxidative Phosphorylation. Vessel-Independent Metabolic Fitness Advantage

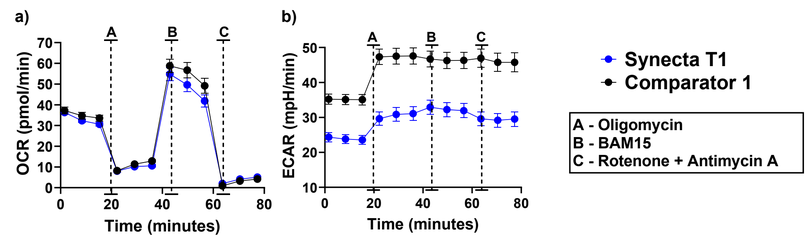


Fig. 3. Metabolic profiling of T cells activated with Synecta T1 or Comparator 1 by mitochondrial stress assay. (A) Oxygen consumption rate (OCR) over time following sequential treatment with oligomycin (A), BAM15 (B), and rotenone plus antimycin A (C). (B) Extracellular acidification rate (ECAR) over time following sequential treatment with oligomycin (A), BAM15 (B), and rotenone plus antimycin A (C).

Taken together, the metabolic data show that Synecta T1 maintains OCR while lowering ECAR relative to Comparator 1, suggesting **reduced glycolytic reliance and a fitter, more memory-associated T cell state**, consistent with the link between metabolic programming and memory development described by Fraietta et al. (2016).

CONCLUSIONS

1. Synecta T1 is fully compatible with the closed and automated IRO platform, enabling Day 0 activation and transduction in a bead-free workflow without soluble cytokines during culture.
2. The Synecta T1 + IRO combination achieved the highest transduction efficiency and total transduced yield across all conditions tested.
3. A 48-hour static phase before mixing further amplified output in IRO, producing the highest Day 7 CAR-T yield and showing reproducible performance across runs.
4. Synecta T1 shifts T cells toward a fitter metabolic state, with lower glycolytic activity and maintained oxidative metabolism.
5. Synecta T1 generates T_{CM}-dominant T cells with balanced CD4/CD8 ratios at Day 7 across vessel formats, supporting consistent product quality.