Improved T-Cell Expansion, Viability, and Metabolic Fitness with Synecta™ Cell-Derived Nanoparticles Across Clinically Relevant Media Conditions

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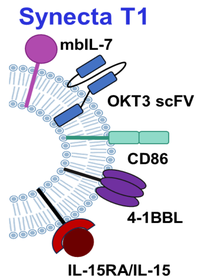
Poster #8

Background

T cell activation quality underpins reproducibility in engineered T-cell manufacturing, affecting expansion, viability, transduction efficiency, and functional fitness. Variability at this step drives batch inconsistency, manufacturing delays, and limited patient access. Conventional bead-based activation induces strong, non-physiologic stimulation, often increasing serum dependence, heterogeneity, and glycolytic stress during prolonged culture. These features contribute to donor variability and manufacturing risk.

Fig. 1



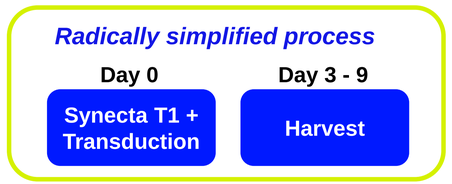
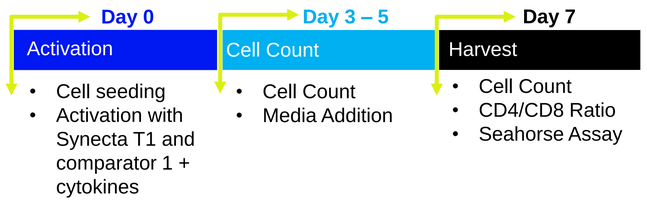


Figure 1. Synecta T1 composition, and possible process of CAR-T manufacturing when using Synecta T1

Synecta is a cell-derived nanoparticle (CDNP) activation platform presenting membrane-bound OKT-3 scFv and co-stimulatory ligands CD86, and 41BBL as well as membrane-bound cytokines IL-7 and IL-15/IL-15Rα in a biologically relevant configuration. This late-breaking analysis evaluates Synecta across xeno-free media systems, including NB-ROC™ and Physiologix™ XF, assessing expansion, viability, and metabolic phenotype. Metabolically efficient activation states have been associated with improved CAR-T persistence and clinical response (Fraietta *et al.*, *Nat Med*, 2018) supporting integrated strategies that improve cellular fitness, accelerated, and scalable manufacturing.

Methods



Media Composition	Media 1	NB-ROC™	Media Supplement	1% GlutaMAX	2% Physiologix™	5% hABs
Media A	X		X	X		
Media A + 2% Physiologix™	X		X	X	X	
NB-ROC™ + 2% Physiologix™		X	X		X	
NB-ROC™ + 5% hABs		X	X			X
Seahorse Media	RPMI 1640 + 10% FBS + 1% HEPES + 1% P/S					

Result

Fig. 2A

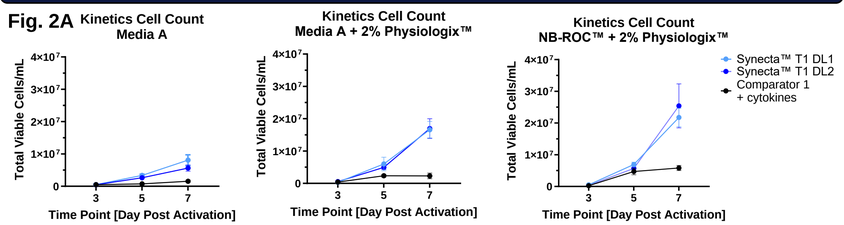


Figure 2A. T cell expansion over time following activation with Synecta T1 or Comparator 1 + cytokines across the indicated culture conditions (Media A, Media A + 2% Physiologix™, and NB-ROC™ + 2% Physiologix™) in healthy donor.

Fig. 2B

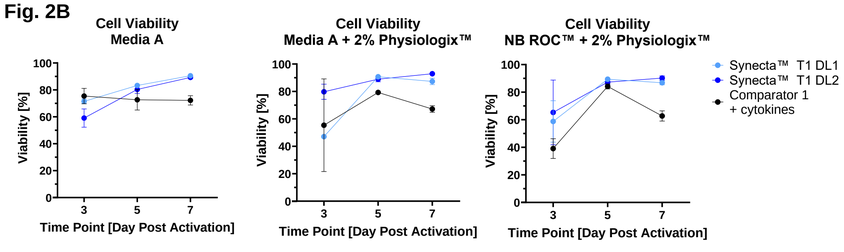


Figure 2B. T cell viability over time following activation with Synecta T1 or Comparator 1 + cytokines across the indicated culture conditions (Media A, Media A + 2% Physiologix™, and NB-ROC™ + 2% Physiologix™) in healthy donor.

Fig. 2C

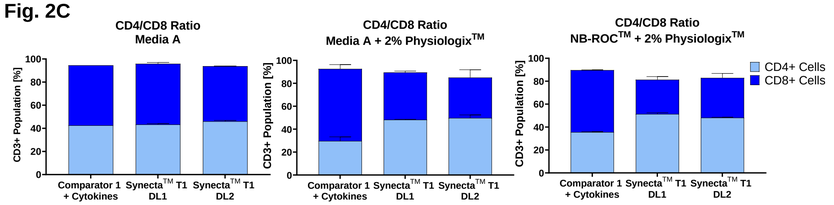


Figure 2C. CD4 and CD8 T-cell composition following activation under different media conditions (Media A, Media A + 2% Physiologix™, and NB-ROC™ + 2% Physiologix™) in healthy donor.. Stacked bars represent the relative proportions of $CD4^+$ and $CD8^+$ T cells at harvest day.

Across conditions, T cells activated with Synecta™ T1 consistently supported superior expansion compared to the comparator, independent of media formulation, with no significant differences observed between DL1 and DL2. The greatest expansion was achieved in cultures maintained in NB-ROC™ supplemented with 2% Physiologix™, highlighting a synergistic effect between Synecta-mediated activation and the optimized nutrient composition of the media. Importantly, enhanced expansion was accompanied by maintained cell viability across all conditions, indicating robust proliferation without evidence of increased cellular stress. CD4/CD8 ratios were comparable in Media A, while an increased proportion of $CD4^+$ T cells was observed in cultures supplemented with 2% Physiologix™.

Result

Fig. 3A

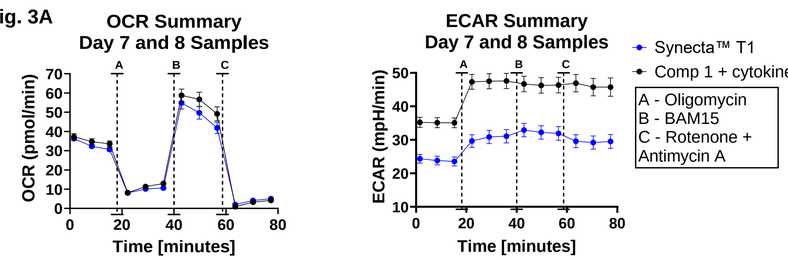


Figure 3A. Oxygen consumption rate (OCR) and extracellular acidification rate (ECAR) measured by Seahorse assay in T cells following activation with Synecta T1 and Comparator 1 + cytokines. (N=4)

Fig. 3B

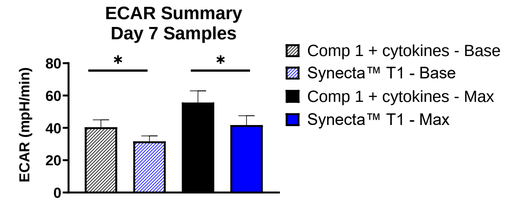


Figure 3B. Summary of ECAR measured by Seahorse XF assay in T cells following activation with Synecta T1 or Comparator 1 plus cytokines, shown relative to baseline and maximal response. (N=3)

Metabolic profiling showed comparable OCR profiles across conditions, while lower ECAR levels were observed in Synecta T1-activated T cells. Despite supporting increased expansion, Synecta activation maintained a balanced glycolytic and oxidative metabolic state, without evidence of excessive glycolytic demand, consistent with a metabolically efficient activation profile.

Conclusions

- **Synecta T1 consistently supported higher T cell expansion** compared to the comparator across all media conditions tested.
- **NB-ROC™ supplemented with 2% Physiologix™ yielded the highest expansion**, highlighting an additive effect between Synecta™-mediated activation and optimized media formulation.
- **Synecta yielded higher cell expansion with sustained cell viability**, indicating robust and consistent performance.
- **Lower reliance on glycolysis is aligned with metabolic programs associated with improved T cell fitness, durability, and functional persistence**, supporting the potential for more sustained therapeutic activity.
- **Collectively, the additive effects of Synecta T1 and NB-ROC™ + Physiologix™ support a mechanistic framework for generating fitter, more persistent T cells, with potential implications for improved patient outcomes in adoptive cell therapy.**

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