

Building manufacturing processes for reproducible, multi-year iPSC-based drug screening



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Background

As high-throughput drug screening (HTS) programs increasingly rely on human induced pluripotent stem cell (iPSC)-derived models, a critical need has emerged: the ability to supply large, relevant models over multi-year screening timelines. This study describes a large-scale manufacturing platform implemented at Ncardia, designed to allow sustained, high-volume, and reproducible cell supply through data-driven process control.

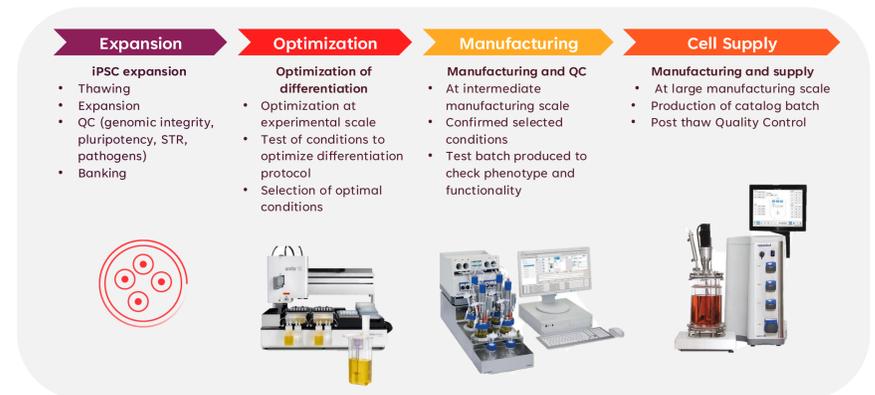
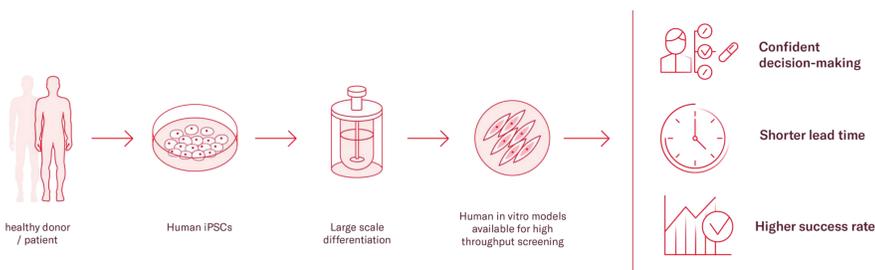


Figure 1. From concept to manufacturing: an end-to-end workflow for scalable production.

The process spans iPSC expansion and banking, small-scale optimization of differentiation conditions, intermediate-scale manufacturing and quality control of differentiated cells, and large-scale manufacturing for cell supply. Optimized conditions are transferred to large-batch production to generate large batches with consistent phenotype and functionality. Iterative monitoring and control of critical process parameters (CPPs), critical material attributes (CMAs), and critical quality attributes (CQAs) ensure reproducibility, batch-to-batch consistency, and reliable downstream performance.

Ncyte® Human Ventricular and Atrial Cardiomyocytes

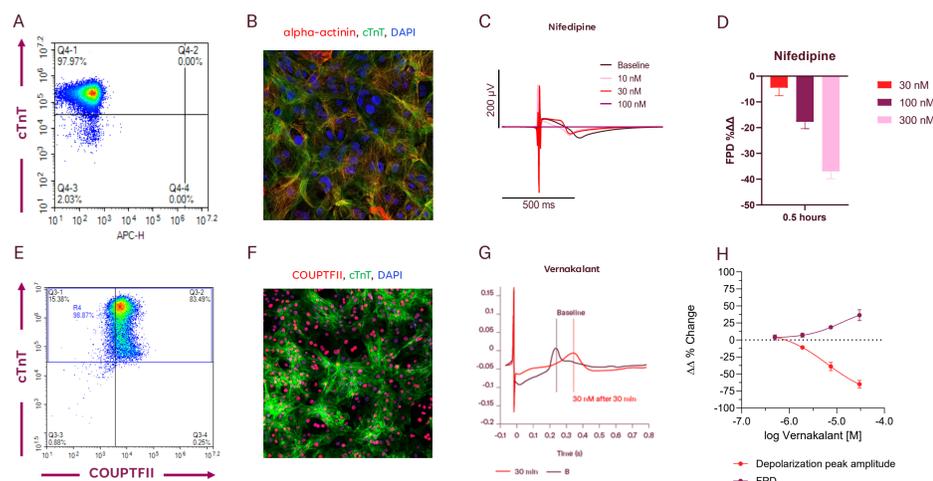


Figure 2. Identity and morphology of Ncyte® human iPSC-derived ventricular- and atrial-like cardiomyocytes. (A-E) Flow cytometry analysis showing high cardiomyocyte purity based on cTnT expression; atrial identity is further distinguished by COUP-TFII expression. (B, F) Immunofluorescence staining of ventricular-like (B) and atrial-like (F) cardiomyocytes demonstrating organized sarcomeric structure (α -actinin or COUP-TFII, cTnT, and DAPI). (C, D) Representative field potential recordings (C) and quantification of field potential duration (FPD; D) following acute nifedipine treatment, showing a dose-dependent electrophysiological response. (G, H) Representative traces (G) and quantification (H) showing dose-dependent effects of vernakalant on depolarization peak amplitude and FPD, consistent with atrial-selective pharmacology. Data are shown as mean \pm SEM.

Ncyte® Heart in a Box™

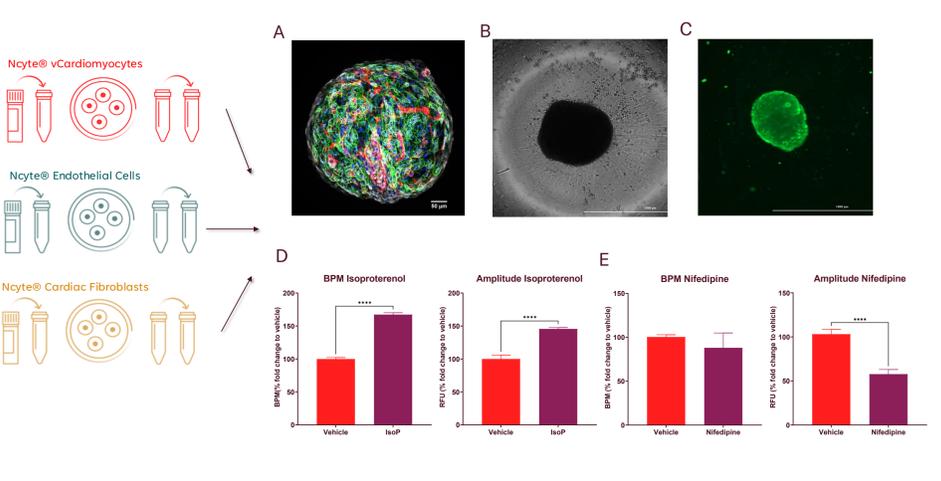


Figure 3. Identity and morphology of Heart in Box™ Microtissue. (A) Immunostaining of Ncyte® Heart in a Box at day 14. Nucleus (blue), Cardiac troponin T (green), CD31 (red), α SMA (white). 20X Image. (B) Brightfield image showing the morphology and structure of a 3D cardiac microtissue, demonstrating the formation of a compact and organized tissue architecture. (C) 3D cardiac microtissues stained using FLIPR6 for calcium imaging. (D) Isoproterenol, a β -adrenergic agonist, significantly increases the beating rate (BPM) and amplitude of 3D cardiac microtissues, indicating a positive chronotropic effect. (E) Nifedipine, a calcium channel blocker, shows no effect on the beating rate (BPM) of 3D cardiac microtissues while significantly decreases the amplitude of contraction in CMs, demonstrating a negative inotropic effect.

Ncyte® NHP-C Cardiomyocytes

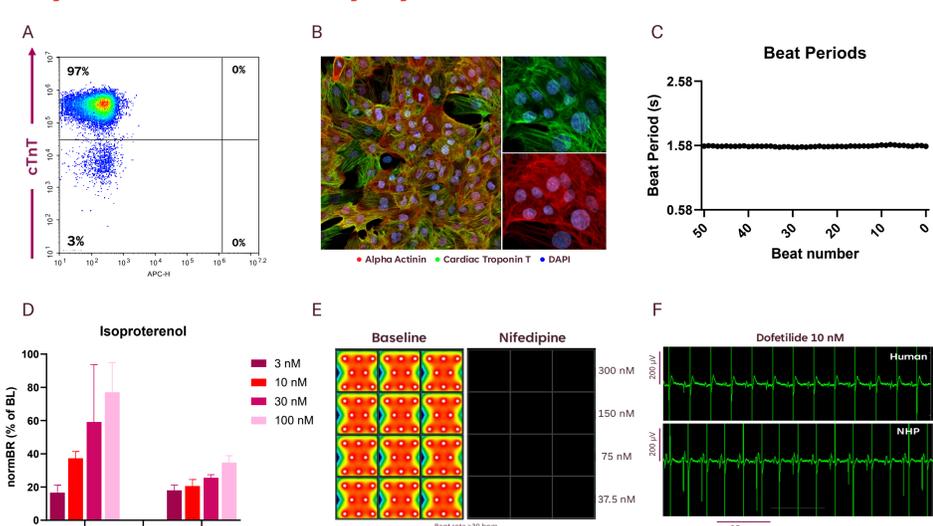


Figure 4. Characterization of the Ncyte® human NHP-C ventricular-like cardiomyocytes. (A) Flow cytometry analysis of cardiac Troponin T (cTnT) expression shows high purity with $\geq 97\%$ cTnT-positive cells. (B) Immunofluorescence staining of NHP-C vCardiomyocytes demonstrating expression of α -Actinin (red), cardiac Troponin T (green), and nuclear counterstain DAPI (blue). (C) Representative beat period recordings of NHP-C vCardiomyocytes show stable beating over successive beats. (D) Response to β -adrenergic isoproterenol, shows a dose dependent increase in beat rate weaker than in human iPSC-derived vCardiomyocytes. (E) Functional response of NHP-C vCardiomyocytes to nifedipine (an L-type calcium channel blocker). Baseline electrical activity is suppressed at all doses (37.5–300 nM). (F) Both human and NHP iPSC-derived vCardiomyocytes exhibit arrhythmic events when exposed to dofetilide.

Ncyte® Microglia

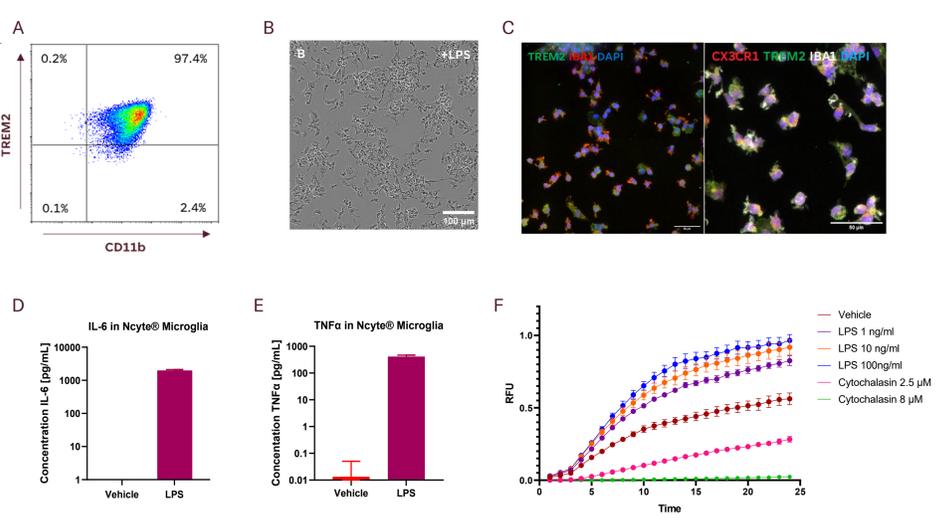


Figure 5. Characterization of iPSC-HSC derived Microglia (A) Flow cytometry analysis of a representative batch of Ncyte® Microglia showing TREM2/CD11b double positive cells (97.4%). (B) bright field image of iPSC-derived microglia after 24-hour exposure to LPS (100 ng/mL). (C) Immunofluorescent staining of Ncyte® Microglia with microglia identity and maturation markers IBA, TREM2, and CX3CR1, co-stained with DNA marker DAPI, on day 7 post-thaw. Scale bar 50 μ m. (D) Release of cytokines IL-6 and TNF α on day 7 post-thaw in Ncyte® Microglia treated for 18hrs with 100ng/mL LPS as measured by Mesoscale Discovery. (E) Representative phagocytosis time-course using pHrodo particles in microglia monocultures treated with vehicle, LPS (1, 10, 100 ng/mL), or cytochalasin D (2.5, 8 μ M). Data are averages \pm SD from n = 4 wells per condition (3 fields of view per well; 20x magnification); time-lapse over 24 h (1 frame/hour).

Conclusions

- Fully controlled stirred-tank bioreactor manufacturing, supported by large-scale process data and analytics, enables a shift from short-term production to sustained, multi-year supply of reproducible iPSC-derived cellular models. Large-batch manufacturing supports consistent phenotype, functionality, and assay performance across extended timelines.
- Standardized yet flexible workflows allow differentiation protocols and quality attributes to be adapted to specific research needs while maintaining robustness and batch-to-batch consistency. Close scientific collaboration ensures that process adjustments are guided by data and aligned with defined performance and quality requirements.
- Our comprehensive suite of cell types has been carefully selected to address diverse research needs. From iPSCs to functional differentiated cells, our portfolio provides the necessary tools to accelerate the discovery of innovative treatments.

The robustness you seek begins with a dependable cell model manufactured at scale

Contact us at support@ncardia.com

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