**Abstract**

The use of stem cells, primary cells and human induced pluripotent stem cells (hiPSCs) has revolutionized the fields of regenerative medicine and cell and gene therapy. These cell populations often require engineering for their generation, reprogramming, differentiation or for targeted gene editing prior to their use. As more robust and scalable cell cultivation methods are developed enabling iPS-Geun/primary cell-derived therapies to advance towards the clinic, it is imperative that researchers consider whether their cell engineering method meets the stringent demands of clinical use - namely the ability to safely and reproducibly modify human primary or stem cells with high efficiency, low cytotoxicity, and at the scale required to treat patients. Flow Electroporation Technology, a non-viral cell engineering technology designed to fulfill these demands, has an established record of rapidly advancing cell- and gene-based therapies to the clinic. In this poster we will demonstrate the high efficiency, high viability, fully scalable engineering of a variety of human primary cells, stem cells and hiPSCs as well as their real-world application in pre-clinical and IND-enabling studies.

**Cardiac “Disease-in-a-Dish” Using iPS-Geun-derived Cardiomyocytes**

CRISPR-Mediated Gene Deletion Recreates Cardiac Arrhythmia

![Diagram](https://example.com/diagram.png)

**20 out of 60 hiPSC clones were positive for deletion using MaxCyte**

**Clinically Relevant Levels of Ex Vivo Gene Correction in X-linked Chronic Granulomatous Disease (X-CGD) Patient Stem Cells**

Gene Editing Moves Towards the Clinic Using Non-viral, Fully Scalable Cell Engineering

![Diagram](https://example.com/diagram.png)

**In vivo results surpassed clinically relevant potency thresholds**

**MaxCyte Flow Electroporation® Technology**

- Ability to engineer primary and stem cells at high efficiency and cell viability enabling their use for:
  - improved, more powerful disease modeling
  - high efficiency gene editing (correction, deletion or insertion)
  - highly potent human therapeutics
- Scalability and regulatory-compliance to rapidly & seamlessly advance from R&D through patient treatment
- Enables previously unfeasible iPS applications
- Improves cell engineering compared to viral- or viral-based delivery methods
  - increased efficiency
  - elimination of safety and toxicity concerns
  - decreased cost and complexity
- Reduced time to clinic
- Designed to meet the stringent demands of cell and gene therapy:
  - highly efficient and reproducible transfection of difficult-to-transfect primary cells
  - non-toxic
  - clinical-scale, regulatory-compliant
- Payload flexibility
- Proven technology supported by numerous publications, clinical trials and 45+ partnered clinical development programs

**References & More Information**


On Demand Webinar: High Transfection Efficiency of Human Induced Pluripotent Stem Cells and Their Derivatives. Presenter: Dr. Nanwei Lin, Ph.D., Chief Scientific Officer, VP, and Co-Founder iXCells Bioengineering. November 2017 [https://example.com/webinar/iXCells-stemcells/]

**Generating an Isogenic Model of Amyotrophic Lateral Sclerosis (ALS)**

Progress Towards Understanding the Molecular Mechanisms of Disease Using Gene Editing

**Figure 1:** Human primary or stem cells were electroporated with pGFP (GFP mRNA for HSCs) using the MaxCyte STX. Transfection efficiency and viability were assessed at 24-48 hours post electroporation.

**Figure 2:** Human iPSCs (iXCells Biotechnologies) were treated with Accutase and dissociated into single cells before electroporation. Cells were electroporated with pGFP using the MaxCyte STX and re-plated on Matrigel-coated plates. Images were taken 24, 48 or 120 hours post electroporation. The transfection efficiency and viability were determined using a Novocye flow cytometer (ACEA). These data were compared to historic results using iXCells previously optimized lipid-based transfection methods.

**Figure 3:** An alamar blue valuation at codon 4 (A4V) of SOD1 causes a rapidly progressive dominant form of amyotrophic lateral sclerosis (ALS) with exclusively motor neuron disease and is responsible for the large majority of SOD1 mutations associated with familial ALS in North America. To create a cellular disease model for ALS, a point mutation (GTC > GCC) was introduced via CRISPR in the normal human SOD1 gene I in hiPSCs. Cells were electroporated with pRNA and repair template and re-plated on Matrigel-coated plates. Single cell colonies were screened using Sanger Sequencing. 3 of 8 clones were positive for the intended mutation: a 37% success rate, which was >10X higher than previously established lipid-based transfection. Wild type and mutant cells were expanded and differentiated into motor neurons using iXCells proprietary methods. Motor neuron markers such as HB9, Choline acetyltransferase (CHAT), Microtubule-associated protein 2 (MAP2) and neurofilament (NF) were expressed and neuronal networks developed by Day 7 for both cell populations.