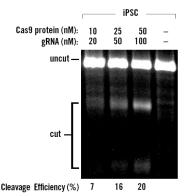


Stem Cell Transfection Solutions

*Trans*IT® Transfection Reagents enable high efficiency transfection of stem cells and other hard to transfect cell types used in stem cell research.

- Perform genome editing with TransIT-X2® Dynamic Delivery System
- Transfect DNA effectively with TransIT®-2020 or TransIT®-LT1 Transfection Reagents
- Perform repeated, low toxicity mRNA transfections using TransIT®-mRNA Transfection Kit
- Electroporate efficiently and cost-effectively with Ingenio® Electroporation Solution



NEW DATA! CRISPR/Cas9 Editing in Human iPSCs

Efficient Genome Editing with CRISPR/Cas9 in Human Induced Pluripotent Stem Cells (iPSCs). The TransIT-X2® Dynamic Delivery System was used to deliver Cas9 protein/guide RNA ribonucleoprotein (RNP) complexes in human induced pluripotent stem cells (iPSCs). For experimental details, please visit: www.mirusbio.com/stemcell

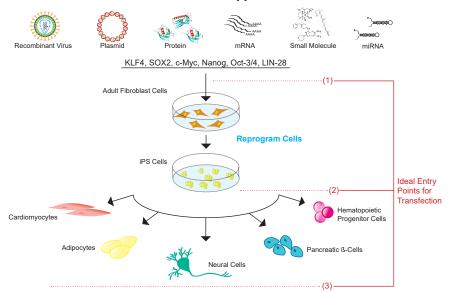




Why Stem Cells?

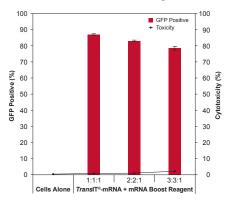
Advances in the field of stem cell differentiation and reprogramming have accelerated drug development by providing disease relevant models for testing. Several of these breakthroughs rely on the use of transfection for non-viral delivery of nucleic acids into different cell types. Mirus Bio provides high efficiency nucleic acid and ribonucleoprotein delivery tools through a suite of *Transl*T® Transfection Reagents and Ingenio® Electroporation Kits that have been validated for many of these applications.

The Role of Transfection in Stem Cells Applications



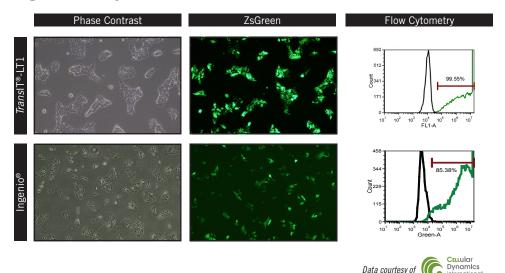
Ideal Entry Points for Transfection in Stem Cell Workflow. Somatic cells such as adult fibroblasts can be transfected or transduced via several methods (e.g. recombinant virus, plasmid, protein, mRNA, small molecule and miRNA) with a combination of transcription factors including KLF4, SOX2, c-Myc, Nanog, Oct-3/4 and LIN-28 to reprogram the cells to a pluripotent state. iPS cells can then be differentiated to a myriad of cell types through growth factor addition and/or transfection of selection markers driven by cell type specific promoters. Stem cell derived cell types such as cardiomyocytes, adipocytes, neural cells, pancreatic- β cells, and hematopoietic progenitor cells can provide researchers with relevant models for their experiments.

Effective, Low Toxicity Delivery of mRNA into Fibroblasts

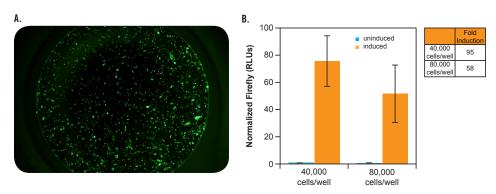


TransIT®-mRNA Provides Effective and Low Toxicity mRNA Transfection. The TransIT®-mRNA Transfection Kit was used to transfect BJ human neonatal foreskin fibroblasts with GFP mRNA incorporating pseudouridine and 5-Me-C modified bases (Trilink Biotechnologies, Inc.). Transfections were performed using 1-3 μl of TransIT®-mRNA Transfection Reagent and mRNA Boost Reagent to deliver 1 μg of RNA (1:1:1, 2:2:1 and 3:3:1; reagent:boost:RNA ratio). At 18 hours post-transfection, GFP was measured and cytoxicity was measured using propidium iodide stain (black line). For experimental details, please visit: www.mirusbio.com/stemcell

High Efficiency DNA Transfection of Human iPS Cells



Ideal Entry Points for Transfection in Stem Cell Workflow. TransIT $^{\circ}$ -LT1 Transfection Reagent was used to reverse transfect 1.3 x 10 $^{\circ}$ iPS cells with a ZsGreen expressing plasmid (Takara Bio USA). Reverse transfections were performed in 6-well plates using 12 µl of TransIT $^{\circ}$ -LT1 Transfection Reagent to deliver 4 µg of DNA (3:1, reagent: DNA). The Ingenio $^{\circ}$ Electroporation Kit was used to transfect 2 x 10 $^{\circ}$ iPS cells on the Amaxa $^{\circ}$ Nucleofector $^{\circ}$ II/2b Device with 8 µg ZsGreen expressing plasmid in 100 µl and plated in 6-well plates at 0.33 x 10 $^{\circ}$ cells/well. Cells were visualized 24 hours post-transfection. Cells were also assayed at 24 hours post-transfection on. The histograms represent the fluorescence intensity of ZsGreen in untransfected cells (black line) compared to cells transfected with plasmid (green line). For experimental details, please visit: www.mirusbio.com/stemcell



Plasmid DNA Delivery to iCell® Cardiomyocytes Using *Trans*IT®-LT1 Transfection Reagent. (A) High efficiency transfection of iCell® Cardiomyocytes (Cellular Dynamics) with a GFP encoding plasmid. Cells were transfected with 100 ng/well of pMAXGFP (Lonza) using *Trans*IT®-LT1 Transfection Reagent with a 2:1 reagent-to-DNA ratio according to the manufacturer's instructions. Fluorescent images were taken 3 days post transfection. (B) cAmp induction measured via a luciferase reporter plasmid. Cells were transfected using *Trans*IT®-LT1 and a CRE-luciferase reporter plasmid. After 18 hours the cAMP pathway was induced using 10 μM isoproterenol for 6 hours. Luciferase activity was measured using the Promega Dual Glo® Luciferase Assay. Data is normalized to the control reporter. For experimental details, please visit: www.mirusbio.com/stemcell







| DNA Transfection | | PRODUCT NO. | QUANTITY |
|-------------------------------------|---|-------------|---------------|
| TransIT-X2® Dynamic Delivery System | | MIR 6003 | 0.3 ml |
| | | MIR 6004 | 0.75 ml |
| | | MIR 6000 | 1.5 ml |
| | | MIR 6005 | 5 x 1 ml |
| | | MIR 6006 | 10 x 1 ml |
| TranslT®-LT1 Transfection Reagent | | MIR 2300 | 1 ml |
| To partie un | | MIR 2304 | 0.4 ml |
| | | MIR 2305 | 5 x 1 ml |
| | | MIR 2306 | 10 x 1 ml |
| TransIT®-2020 Transfection Reagent | | MIR 5400 | 1 ml |
| Senterine Senterine | | MIR 5404 | 0.4 ml |
| | | MIR 5405 | 5 x 1 ml |
| | | MIR 5406 | 10 x 1 ml |
| mRNA Transfection | | PRODUCT NO. | QUANTITY |
| TransIT®-mRNA Transfection Kit | | MIR 2250 | 1 ml |
| | | MIR 2225 | 0.4 ml |
| | | MIR 2255 | 5 x 1 ml |
| | | MIR 2256 | 10 x 1 ml |
| Electroporation | | PRODUCT NO. | QUANTITY |
| Ingenio® Electroporation Kit | lectroporation Kit | | 25 reactions |
| | Compatible with Amaxa® Nucleofactor Device (solution, 0.2 cm cuvettes and cell droppers) | MIR 50115 | 50 reactions |
| | | MIR 50118 | 100 reactions |
| Ingenio® Electroporation Kit | | MIR 50113 | 25 reactions |
| | Compatible with Bio-Rad® and Harvard-BTX® electroporation systems (solution, 0.4 cm cuvettes and cell droppers) | MIR 50116 | 50 reactions |
| | | MIR 50119 | 100 reactions |
| | Ingenio® Electroporation Solution | | 25 reactions |
| ngenio® Electroporation Solu | ition | MIR 50111 | 20 1000010110 |
| Ingenio® Electroporation Solu | rtion | MIR 50114 | 50 reactions |



