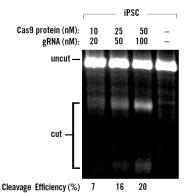


Stem Cell Transfection Solutions

*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 *Trans*IT-X2® Dynamic Delivery System
- Transfect DNA effectively with *Trans*IT®-2020 or *Trans*IT®-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).



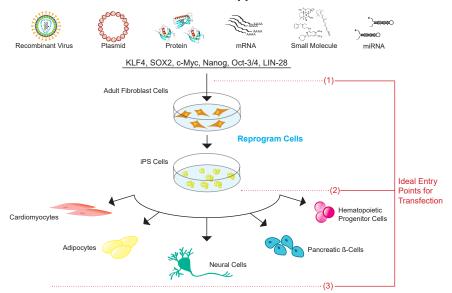




Why Stem Cells?

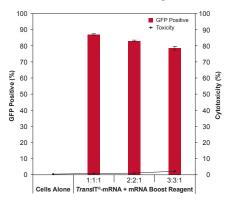
Advances in the field of stem cell differentiation and reprogramming have accelerated drug development by providing disease relevant models for testing. Several of these breakthroughs rely on the use of transfection for non-viral delivery of nucleic acids into different cell types. Mirus Bio provides 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



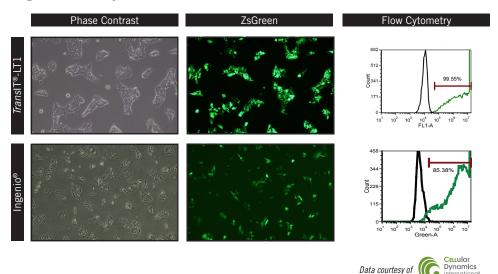
Ideal Entry Points for Transfection in Stem Cell Workflow. Somatic cells such as adult fibroblasts can be transfected or transduced via several methods (e.g. recombinant virus, plasmid, protein, mRNA, small molecule and miRNA) with a combination of transcription factors including KLF4, SOX2, c-Myc, Nanog, 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

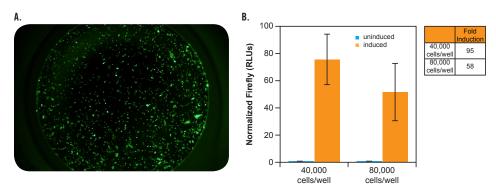


TransIT®-mRNA Provides Effective and Low Toxicity mRNA Transfection. The TransIT®-mRNA Transfection Kit was used to transfect BJ human neonatal foreskin fibroblasts with GFP mRNA incorporating pseudouridine and 5-Me-C modified bases (Trilink Biotechnologies, Inc.). Transfections were performed using 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).

High Efficiency DNA Transfection of Human iPS Cells



Ideal Entry Points for Transfection in Stem Cell Workflow. TransIT®-LT1 Transfection Reagent was used to reverse transfect 1.3 x 106 iPS cells with a ZsGreen expressing plasmid (Takara Bio USA). Reverse transfections were performed in 6-well plates using 12 μ l of TransIT®-LT1 Transfection Reagent to deliver 4 μ g of DNA (3:1, reagent: DNA). The Ingenio® Electroporation Kit was used to transfect 2 x 106 iPS cells on the Amaxa® Nucleofector® II/2b Device with 8 μ g ZsGreen expressing plasmid in 100 μ l and plated in 6-well plates at 0.33 x 106 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).



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.









Broad Spectrum DNA Transfection		PRODUCT NO.	QUANTITY
TransIT-X2® Dynamic Delivery System		MIR6003	0.3 ml
		MIR6004	0.75 ml
		MIR6000	1.5 ml
		MIR6005	5 x 1 ml
		MIR6006	10 x 1 ml
TransIT®-LT1 Transfection Reagent		MIR2300	1 ml
The second secon		MIR2304	0.4 ml
		MIR2305	5 x 1 ml
		MIR2306	10 x 1 ml
TransIT®-2020 Transfection Reagent		MIR5400	1 ml
To Marie		MIR5404	0.4 ml
		MIR5405	5 x 1 ml
		MIR5406	10 x 1 ml
mRNA Transfection		PRODUCT NO.	QUANTITY
TransIT®-mRNA Transfection Kit		MIR2250	1 ml
		MIR2225	0.4 ml
		MIR2255	5 x 1 ml
		MIR2256	10 x 1 ml
Electroporation		PRODUCT NO.	QUANTITY
Ingenio® Electroporation Kit	Compatible with Amaxa® Nucleofactor Device (solution, 0.2 cm cuvettes and cell droppers)	MIR50112	25 reactions
		MIR50115	50 reactions
		MIR50118	100 reactions
Ingenio® Electroporation Kit	Compatible with Bio-Rad® and Harvard-BTX® electroporation systems (solution, 0.4 cm cuvettes and cell droppers)	MIR50113	25 reactions
100		MIR50116	50 reactions
		MIR50119	100 reactions
Ingenio® Electroporation Solution		MIR50111	25 reactions
Marie Control of the		MIR50114	50 reactions
		MIR50117	100 reactions

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