Version Date: 1/12/2025

Product Summary

Human iPS Cell Complete Reprogramming Kit

Catalog Number: MR1003-K

Product Overview	
Product Name	**Human iPS Cell Complete Reprogramming Kit**
Catalog #	MR1003-K
Product Form(s)	Liquid base medias Frozen cell culture supplements
Cell Species	Human

Product Overview

Specifically designed to reprogram starting cell colonies safely and efficiently. This kit can be used with primary fibroblast or peripheral blood mononuclear cells (PBMCs) such as lymphocytes, monocytes, and dendritic cells. Starting cells can be adapted for adherent growth or suspension-based applications, as our protocol is adapted for traditional electroporation or chemically mediated transfection, depending on customer transduction preference.

Our proprietary method requires a single transfection step and uses episomal DNA and small molecule inhibitors to efficiently reprogram primary cells and maintain pluripotency markers. Target cells at low passage numbers that are mitotically active are highly recommended.

Important: This product can be used under Biosafety Level 1 (BL-1) containment with biological safety cabinet and laminar flow hood, and with appropriate personal safety equipment to prevent mucosal exposure/splash.

Base medias are shipped with gel packs. Episomal reprogramming mix and required supplements are shipped on dry ice.

Note: This product has been tested using Cellular Engineering Technologies Inc. ("CET") Human Foreskin Fibroblast iPS Cells CR1001-500 Human Foreskin Fibroblast iPS Cells, CR1002-500 Human Multipotent iPS Cells, CR1003-500 Human CD34+ iPS Cells, and CR1018-500 Human Amniotic Membrane iPS Cells (all not included). Although investigators are welcome to use this product with other cell products, CET cannot guarantee this product's performance. Please refer to CET's Terms & Conditions, available at www.cet.bio.

Product Image

Included Components:

Human iPSC Reprogramming Base Media (250mL)
Human iPSC Reprogramming Supplement (495µL)
Human iPSC Episomal DNA Mix (Cat. MR1004)
Human iPSC Growth Media Kit (Cat. MR1001-K)
- Human iPSC Growth Base Media (500mL)
- Human iPSC Growth Media Supplement (7mL)

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Overview of iPSC Reprogramming

Overview of iPSC Reprogramming	
Induced Pluripotent Stem Cells (iPSCs)	Induced pluripotent stem cells (iPSCs) are genetically reprogrammed adult somatic cells through genetic reprogramming. This process traditionally involves introducing specific transcription factors, such as *Oct4*, *Sox2*, *Klf4,* and *c-Myc* or *l-Myc*, to revert the mature cells into an embryonic-like pluripotent state. iPSCs can differentiate into any cell type of the body and hold great potential for regenerative medicine, disease modeling, and drug discovery. iPSC reprogramming methods have advanced significantly, yet several challenges have persisted, which present challenges for the researchers and therapeutic developers: • **Safety Concerns**: Reprogramming can introduce genetic and epigenetic abnormalities, potentially leading to tumorigenicity. Ensuring genomic stability during and after reprogramming is crucial to mitigate these risks.[1] • **Low Efficiency**: The process of converting somatic cells into iPSCs has remained inefficient, often yielding a small percentage of successfully reprogrammed cells. This inefficiency necessitates the culture of large numbers of cells, making the process of iPSC generation labor-intensive and costly.[2] • **Immunogenicity**: iPSCs may elicit immune responses, even when derived from the same individual, complicating their therapeutic application. Address this requires strategies to reduce or eliminate immunogenic factors. • **Incomplete Reprogramming**: Achieving full pluripotency is challenging, with some cells exhibiting partial reprogramming, leading to variability in differentiation potential and function.[3] • **Transfection Methods**: The choice of transfection method impacts reprogramming efficiency and safety. Integrating viral vectors pose risks of insertional mutagenesis, while non-integrating methods may have lower efficiency.[4]
CET's Reprogramming Method	CET's patented proprietary method uses non-integrating, episomal DNA and small molecule inhibitors that reprogram starting cell colonies safely and efficiently. Unlike other reprogramming techniques, which require multiple transfections and 3-4 weeks to clear delivery vectors, our technology requires a single transfection step and delivers reprogrammed colonies in under 20 days with no karyotypic abnormalities. This kit does not use *c-Myc* and *Lin28* transcription factors, which have traditionally governed cell reprogramming efficiency levels. This method can be used to reprogram primary fibroblast or peripheral blood mononuclear cells (PBMCs) such as lymphocytes, monocytes, and dendritic cells. Starting cells can be adapted for adherent growth or suspension-based applications, and our protocols can be adapted for traditional electroporation or chemically mediated transfection, depending on your preferred method. Episomal vectors provide an efficient alternative to integrative viral and non-viral delivery systems, and before, and present significantly lower risk for insertional mutagenesis and innate immune response. Due to their transient nature, episomal DNA is lost from cells at a rate of 5% per cell generation, hence episomal-free iPSC can typically be harvested by day 15 from initial transduction.
Overview of Episomal DNA plasmids	Episomal DNA vectors are genetic tools used in molecular biology and biotechnology for the delivery and expression of genes in target cells. They are distinct from chromosomal DNA because they replicate independently of the host genome, existing as extrachromosomal circular DNA molecules. This feature makes them versatile for various applications, particularly in gene therapy, protein production, and research on gene function. Key characteristics of episomal DNA vectors are the following: 1. **Autonomous Replication**: Episomal vectors can replicate independently in host cells

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Description of the Reprogramming System

	2. **Non-integration**: Episomal vectors do not integrate into the host genome, reducing risk of insertional mutagenesis. 3. **Stability**: Episomal vectors are stable depending on the type of origin of replication and selective pressure applied during cell culturing.
Description of the System	
CET Episomal Vector Design and Small Molecule Inhibitors	Some of the key features of CET's episomal DNA vectors are the following: • An pCEP4 vector backbone with a CMV promoter and a hygromycin resistance marker to promote high-level expression in large mammalian models. • An Epstein-Barr Virus (EBV) origin of replication (OriP) is a well-characterized sequence derived from the viral genome that allows episomal maintenance and replication of vectors and enables long-term retention of the vector in proliferating cells without integration into the host genome. • A human CMV intermediate script to promote high-level expression. • A small molecule inhibitor that suppressed p53 expression to inhibit cell cycle arrest and cell apoptosis • A small molecule inhibitor to promote glycolysis
Advantages of the Human iPSC Complete Reprogramming Kit	• Elevated levels of cell reprogramming efficiency for traditionally challenging blood-derived cell types (e.g., $CD34^+$) • Reprogramming protocol requires a single transduction step to deliver episomal plasmid vectors—multiple or daily transfections not required • Robust iPSC colonies from patient-derived or reference mononuclear cells in approximately 20 days • Non-viral, episomal DNA vectors safely clear in approximately 20 days versus weeks to months for persistent viral vectors • Cost-effective and simple protocol reduces hands-on time and consumable usage

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Reprogramming CD34⁺ Cells

Required Materials	
Cells and vectors	• Peripheral blood mononuclear cells (PBMCs) to reprogram (not included) • Human iPSC Episomal Reprogramming Mix (~50ug) (Cat. MR1004) for delivery of exogenous reprogramming vectors. *Note: CET strongly recommends using PBMCs extracted from fresh blood by a conventional method (i.e., Ficoll-Paque purification) or frozen PBMCs.*
Media and consumables	• PBMC media (not included) for Day -7 through Day 0 cell line expansion • CET Human iPSC Reprogramming Base Media (250mL) (included) for plating of transduced cells from Day 3 to Day 14 • CET Human iPSC Reprogramming Supplement (495µL) (included) for activation of base reprogramming media • CET Human iPSC Growth Media Kit (Cat. MR1001-K) (included) for expansion of transduced cells on extracellular matrix from Day 14 and thereafter, live staining and picking of iPSC colonies. *Items not included:* • **Extracellular Matrix** - Basement membrane matrix suitable for adherent cell culture. In-house recommendation Corning® Matrigel® hESC-Qualified Matrix, LDEV-Free (Cat. 354277) • **Penicillin/Streptomycin/Amphotericin B solution or Penicillin/Streptomycin** solution, 100x[5] ("antibiotic/antimycotic solution") • **Surface Disinfectant** solution containing 70% isopropanol and 30% sterile water recommended
Recommended Equipment	**Electroporation Platform** – This reprogramming protocol recommends electroporation to deliver episomal DNA plasmids across the cell membranes of target cells. We have observed higher post-transfection cell viabilities and the ability to deliver larger genetic payloads (> 7kb) when reprogramming challenging, suspension-based cell samples. Our recommended electroporation platform is the Neon™ NxT Electroporation System manufactured by Thermo Fisher. Users must be proficient when using this recommended system or a comparable electroporation system of their choice. Please refer to the manufacturer's user guide and required materials list when preparing to run this reprogramming protocol. Please refer to the Neon NxT Electroporation System User Guide (Pub. No. MAN0026677 B.0) for reference. *Note: Chemical methods such as cationic lipid transfection can also be used to deliver genetic payloads, however, we have observed that this method is challenging for suspension-based cell samples.*

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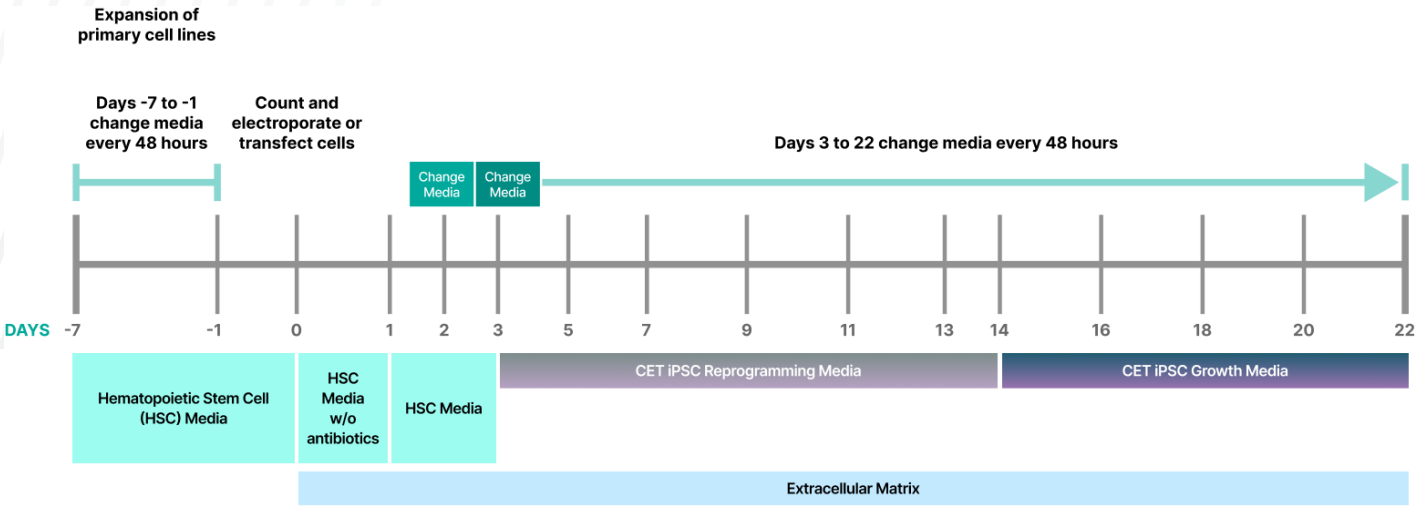
Reprogramming CD34⁺ Cells

Simplified Workflow



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Reprogramming CD34+ Cells

Media Formulation Instructions – Human iPSC Reprogramming Media	
Defrosting and Preparation	1. Defrost the Human iPS Growth Supplement at 4°C twenty-four (24) hours before the media is to be prepared in a 37°C water bath until ice in the tubes is no longer visible. Never defrost the iPS Growth Supplement using a 37°C water bath. It is normal for Human iPS Growth Supplement to appear hazy or have suspended solutes. 2. Gently mix by inversion. 3. Immediately disinfect the tubes and the bottle containing the Human iPS Growth Base Media with 70% isopropanol.
Mixing	4. Working in a laminar flow hood, remove 12 mL of the Human iPSC Growth Base Media from the bottle and discard it. This and all other procedures must be done in a sterile manner. 5. Add the complete contents of the Human iPSC Growth Supplement to the Human iPSC Growth Base Media. 6. Cap the bottle containing the mixed liquid solution and gently swirl it a few times. This formulated media is now considered **complete iPSC Growth Media** and ready to use with cells.

Media Formulation Instructions – Human iPSC Growth Media	
Defrosting and Preparation	1. Defrost Human iPSC Reprogramming Supplement at 4°C twenty-four hours before the media is to be prepared and 2.5mL of antibiotic/antimycotic solution (not included) in a 37°C water bath until ice in the tubes is no longer visible. Never defrost the iPS Reprogramming Supplement using a 37°C water bath. 2. Immediately disinfect the tubes and the bottle containing the Human iPS Reprogramming Base Media with 70% isopropanol (not included).
Mixing	3. Working in a laminar flow hood, remove 2.5 mL of the Human iPSC Reprogramming Base Media from the bottle and discard it. This and all other procedures must be done in a sterile manner. 4. Add the complete contents of the Human iPSC Reprogramming Supplement to the Human iPSC Base Media. 5. Add 2.5mL of the antibiotic/antimycotic solution to the reprogramming base media[1]. 6. Cap the bottle containing the mixed liquid solution and gently swirl it a few times. This formulated media is now considered **complete iPSC Reprogramming Media** and ready to use with cells.

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Reprogramming CD34+ Cells

Protocol for Reprogramming CD34+ Cells	
Day -7	**Seed PBMCs** *(general guidelines below; please refer to vendor cell culturing instructions for PBMCs)*
	1. Seven days before planned transduction, remove vial(s) of PBMCs from liquid nitrogen storage. 2. Defrost the vial(s) of cells in a 37°C water bath with constant, moderate agitation until ice in the ampoule is barely visible. DO NOT OVERTHAW. 3. Spray the outside of the vial with 70% isopropanol before placing it in the cell culture hood. 4. Gently transfer the PBMCs into a 15 mL conical tube. Slowly (dropwise) add 5 – 10 mL pre-warmed complete PBMC medium (see vendor instructions) to the cell suspension. 5. Remove an aliquot of cells to count and determine cell viability. 6. Centrifuge the cell suspension at 200 x g for 10 minutes, discard the supernatant, and resuspend the cells in complete PBMC medium to 5 x 10^5 cells / mL
Day -3 to -1	**Observe cells and add fresh PBMC media**
	7. Continue to expand PBMCs to a target density of 1 x 10^6 cells / mL. 8. Observe confluency. We recommend about 30-60% confluency on the day of transduction. Because over-confluency results in decreased transduction efficiency, we recommend replating your cells to achieve 30-60% confluency if your cells have become over confluent during culturing. 9. For each suspension cell line to be transduced, coat one (1) well of a tissue culture dish with selected extracellular matrix *Note: Twenty-four (24) hours before transduction, PBMC growth media* ***cannot contain antibiotics and/or antifungals.***
Day 0	**Count cells and perform transduction**
	11. Count and spin cells to pellet at 200 X G for five (5) minutes in a swing bucket rotor. Cell density should be 1 x 10^6 cells / mL. 12. Re-suspend the cell pellet in one hundred (100) µL (microliters) of Neon Electroporation Buffer S. 13. Add twelve (12) µL (microliters) of **Human iPSC Episomal Reprogramming Mix** (Cat. MR1004) to this tube. This represents approximately ten (10) µg (micrograms) of DNA. 14. Mix gently using a micropipettor 15. Using a Neon Electroporation Tip-100 (not included), introduce cells and episomal DNA. 16. Using Neon Electroporation Buffer E2 (not included) for the chamber buffer, electroporate cells at 1650 V for ten (10) milliseconds for three (3) cycles. Immediately after electroporation, place cells in your **complete iPSC Growth Media** on the coated 6-well dish. ***Note: This media must NOT contain ANTIBIOTICS and/or ANTIFUNGALS for the first 24 hours following transduction.***
Days 1-3	**First media change post-transduction**
	17. At the end of 24 hours, gently tilt the plate so cells settle to the bottom. 18. Note: It will take approximately 72 hours post-electroporation for suspension cells to settle and adhere. **It is critical not to aspirate suspension cells while conducting a media change.** 19. Using a serological pipette, gently withdraw one-half of the volume of the antibiotic/antifungal-free media. Replace with a half volume of **complete iPSC Reprogramming Media**. 20. Observe iPSC colony emergence *Note: Although exogenous gene expression should start within 12 hours, robust gene expression can be detected by RFP fluorescence after 48 hours post-transduction.*
Days 3-14	**iPSC Morphology Emergence and complete iPSC Reprogramming Media changes**

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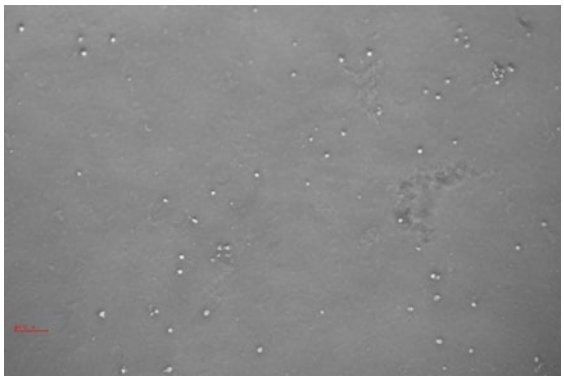
	21. After 72 hours following transduction, complete a full media replacement with **complete iPSC Reprogramming Media**. 22. After this complete iPSC Reprogramming Media replacement, perform a full replacement of **complete IPSC Reprogramming Media** every 48 hours. Suspension cells should be adherent by this time. All cells should start becoming more cuboidal or epithelial in appearance and start forming small putative iPSC colonies by the end of 14 days.
Days 14+	**iPSC Colony Emergence and Transition to complete iPSC Growth Media**
	23. Perform a full media replacement with **complete iPSC Growth Media** every 24 hours. Although it is difficult to predict when mature iPSC colonies will emerge, this process should take approximately 17 days post-electroporation. Monitor iPSC colonies daily. iPSC colonies are ready to be passaged when they have sharp, distinct edges. 24. For passaging and growth directions, please refer to CET's guide on growing and maintaining iPSC colonies. *Note:* CET highly recommends the usage of antibiotic and antimycotics for long-term cell culturing.

Expected Visual Results

iPSC morphology emergence from reprogrammed CD34⁺ cells	
Day 2 post-transduction Reprogrammed cells recovering from transduction	
Day 10 post-transduction Morphology changes indicating reprogramming	

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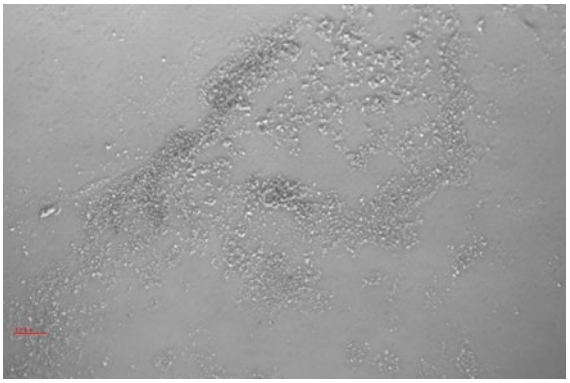
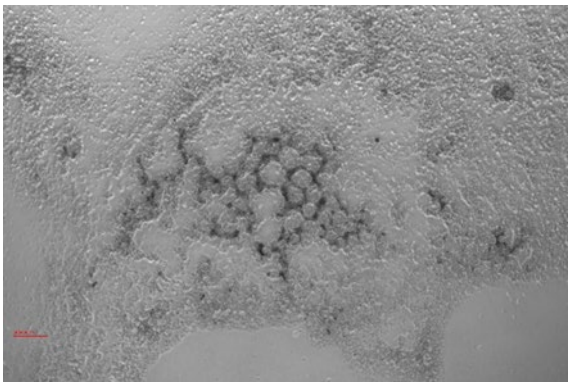
Reprogramming CD34+ Cells

Day 14 post-transduction Continued morphology changes and colony formation	
Day 17 post-transduction iPSC colonies are formed and ready for colony picking	

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Reprogramming Fibroblast Cells

Required Materials	
Cells and vectors	• Human fibroblast cells (HFs) to reprogram (not included) • Human iPSC Episomal Reprogramming Mix (~50ug) (Cat. MR1004) for delivery of exogenous reprogramming vectors.
Media and consumables	• HF expansion media (not included) for Day -7 through Day 0 cell line expansion • CET Human iPSC Reprogramming Base Media (250mL) (included) for plating of transduced cells from Day 3 to Day 14 • CET Human iPSC Reprogramming Supplement (495µL) (included) for activation of base reprogramming media • CET Human iPSC Growth Media Kit (Cat. MR1001-K) (included) for expansion of transduced cells on extracellular matrix from Day 14 and thereafter, live staining and picking of iPSC colonies. *Items not included:* • **Extracellular Matrix** - Basement membrane matrix suitable for starting adherent cell culture. In-house recommendation Geltrex™ hESC-Qualified, Ready-To-Use, Reduced Growth Factor Basement Membrane Matrix (Cat. A1569601) • **Phosphate-buffered saline (PBS)** - based on customer preference, we recommend using Phosphate-buffered saline (DPBS, 1X), Dulbecco's formula (Cat. J67670.AP) manufactured by Thermo Fisher • **Trypsin-EDTA (0.25%)** - based on customer preference • **Penicillin/Streptomycin/Amphotericin B solution or Penicillin/Streptomycin** solution, 100x[6] ("antibiotic/antimycotic solution") • **Surface Disinfectant** solution containing 70% isopropanol and 30% sterile water recommended
Recommended Equipment	**Chemical Transfection Platform** – This reprogramming protocol recommends chemical transfection to deliver episomal DNA plasmids across the cell membranes of target cells. We have observed higher post-transfection cell viabilities and the ability to deliver larger genetic payloads (> 7kb) when reprogramming adherent-based cell samples. Our recommended chemical transfection platform is the Lipofectamine™ 3000 Transfection Reagent manufactured by Thermo Fisher. Users must be proficient when using this recommended system or a comparable chemical transfection system of their choice. Please refer to the manufacturer's user guide and required materials list when preparing to run this reprogramming protocol.

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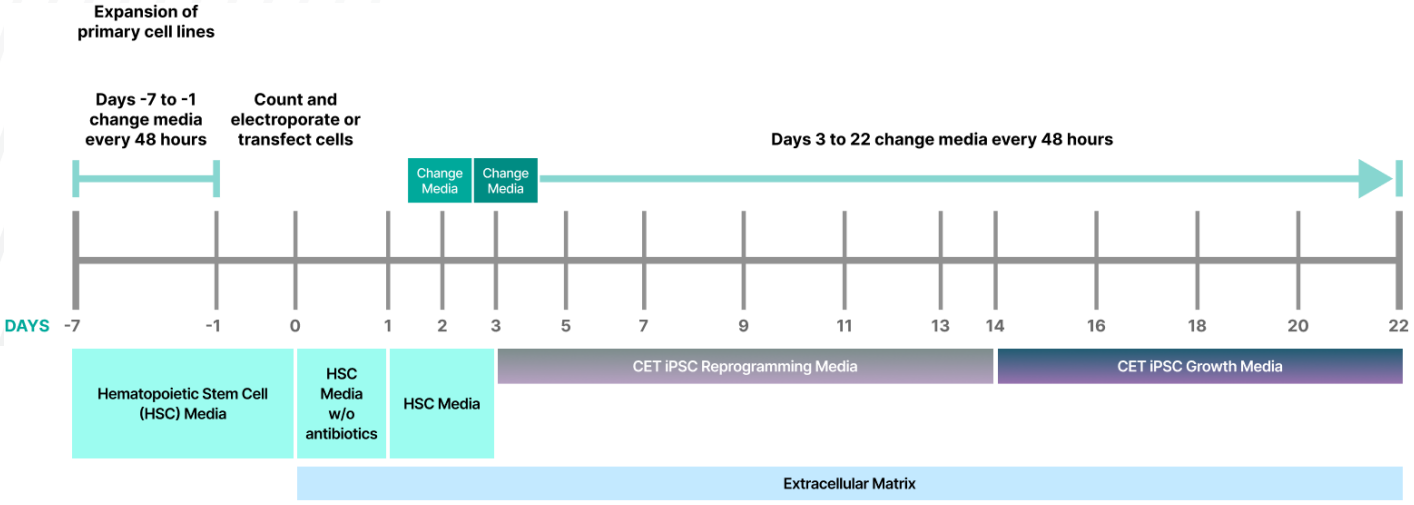
Reprogramming Fibroblast Cells

Simplified Workflow



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Reprogramming Fibroblast Cells

Media Formulation Instructions – Human iPSC Reprogramming Media	
Defrosting and Preparation	1. Defrost the Human iPS Growth Supplement at 4°C twenty-four (24) hours before the media is to be prepared in a 37°C water bath until ice in the tubes is no longer visible. Never defrost the iPS Growth Supplement using a 37°C water bath. It is normal for Human iPS Growth Supplement to appear hazy or have suspended solutes. 2. Gently mix by inversion. 3. Immediately disinfect the tubes and the bottle containing the Human iPS Growth Base Media with 70% isopropanol.
Mixing	4. Working in a laminar flow hood, remove 12 mL of the Human iPSC Growth Base Media from the bottle and discard it. This and all other procedures must be done in a sterile manner. 5. Add the complete contents of the Human iPSC Growth Supplement to the Human iPSC Growth Base Media. 6. Cap the bottle containing the mixed liquid solution and gently swirl it a few times. This formulated media is now considered **complete iPSC Growth Media** and ready to use with cells.

Media Formulation Instructions – Human iPSC Growth Media	
Defrosting and Preparation	1. Defrost Human iPSC Reprogramming Supplement at 4°C twenty-four hours before the media is to be prepared and 2.5mL of antibiotic/antimycotic solution (not included) in a 37°C water bath until ice in the tubes is no longer visible. Never defrost the iPS Reprogramming Supplement using a 37°C water bath. 2. Immediately disinfect the tubes and the bottle containing the Human iPS Reprogramming Base Media with 70% isopropanol (not included).
Mixing	3. Working in a laminar flow hood, remove 2.5 mL of the Human iPSC Reprogramming Base Media from the bottle and discard it. This and all other procedures must be done in a sterile manner. 4. Add the complete contents of the Human iPSC Reprogramming Supplement to the Human iPSC Base Media. 5. Add 2.5mL of the antibiotic/antimycotic solution to the reprogramming base media[1]. 6. Cap the bottle containing the mixed liquid solution and gently swirl it a few times. This formulated media is now considered **complete iPSC Reprogramming Media** and ready to use with cells.

Protocol for Reprogramming Fibroblast Cells	
Day -7	**Seed HFs** *(general guidelines below; please refer to vendor cell culturing instructions for HFs)*
	1. Seven days before planned transduction, remove vial(s) of HFs from liquid nitrogen storage. 2. Follow vendor directions for thawing and plating HF cells 3. Regularly remove an aliquot of cells to count and determine cell viability.
Day -3 to -1	**Observe cells and add fresh HF media**
	4. Continue to expand HFs to a target density of 1 x 10^6 cells / mL. 5. Observe confluency. We recommend about 30-60% confluency on the day of transduction. Because over-confluency results in decreased transduction efficiency, we recommend replating your cells to achieve 30-60% confluency if your cells have become over confluent during culturing.

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	6. For each cell line to be transduced, coat one (1) well of a tissue culture dish with selected extracellular matrix. Please follow specific instructions for coating cell culture surfaces with selected extracellular matrix. *Note: Twenty-four (24) hours before transduction, HF growth media* ***cannot contain antibiotics and/or antifungals.***
Day 0	**Count cells and perform transduction**
	17. Count and spin cells to pellet at 200 X G for five (5) minutes in a swing bucket rotor. Cell density should be 1 x 10^6 cells / mL. 18. Follow transfection reagent manufacturer's instructions for introducing one hundred (100) µL of CET Human iPSC Episomal Reprogramming Mix to cell suspension, this represents approximately ten (10) µg (micrograms) of DNA. 19. Follow transfection reagent manufacturer's instructions for introducing **complete iPSC Growth Media** on coated cell culture surfaces. ***Note: This media must NOT contain ANTIBIOTICS and/or ANTIFUNGALS for the first 24 hours following transduction.***
Days 1-3	**First media change post-transduction**
	23. At the end of 24 hours, gently tilt the plate so cells settle to the bottom. 24. Note: It will take approximately 72 hours post-electroporation for suspension cells to settle and adhere. **It is critical not to aspirate suspension cells while conducting a media change.** 25. Using a serological pipette, gently withdraw one-half of the volume of the antibiotic/antifungal-free media. Replace with a half volume of **complete iPSC Reprogramming Media**. 26. Observe iPSC colony emergence *Note: Although exogenous gene expression should start within 12 hours, robust gene expression can be detected by RFP fluorescence after 48 hours post-transduction.*
Days 3-14	**iPSC Morphology Emergence and complete iPSC Reprogramming Media changes**
	27. After 72 hours following transduction, complete a full media replacement with **complete iPSC Reprogramming Media**. 28. After this complete iPSC Reprogramming Media replacement, perform a full replacement of **complete IPSC Reprogramming Media** every 48 hours. Suspension cells should be adherent by this time. All cells should start becoming more cuboidal or epithelial in appearance and start forming small putative iPSC colonies by the end of 14 days.
Days 14+	**iPSC Colony Emergence and Transition to complete iPSC Growth Media**
	25. Perform a full media replacement with **complete iPSC Growth Media** every 24 hours. Although it is difficult to predict when mature iPSC colonies will emerge, this process should take approximately 17 days post-electroporation. Monitor iPSC colonies daily. iPSC colonies are ready to be passaged when they have sharp, distinct edges. 26. For passaging and growth directions, please refer to CET's guide on growing and maintaining iPSC colonies. *Note:* CET highly recommends the usage of antibiotic and antimycotics for long-term cell culturing.

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Kit Components and Storage Requirements

Storage and Stability			
	Volume	**Storage Temperature**	**Storage Time**
Human iPSC Complete Reprogramming Kit Cat. MR1003-K			
iPSC Reprogramming Base Media	250 mL	4°C	3 months
iPSC Reprogramming Supplement		-20°C	3 months
complete iPSC Reprogramming Media *(see Media Formulation Instructions)*		4°C	Not applicable (use entire contents)
Human iPSC Growth Media Kit Cat. MR1001-K			
iPSC Growth Base Media	500 mL	4°C	3 months
iPSC Growth Media Supplement	7 mL	-20°C	6 months
complete iPSC Growth Media *(see Media Formulation Instructions)*		4°C	Not applicable s(use entire contents)
Human iPSC Episomal Reprogramming Mix Cat. MR1004	50 µg	-20°C	6 months
Avoid repeated exposure to room temperature and light.			

Please contact us at https://cet.bio/contact for the latest service and support information or email us directly at support@cet.bio. Our support team is available to answer your technical questions.

Note: For product user guides and safety data sheets for third-party reagents and other inputs please contact the manufacturer.

[1] Moy AB. The Challenges to Advancing Induced Pluripotent Stem Cell-Depending Cell Replacement Therapy. Med Res Arch. 2023 Nov; 11(11): 4784. DOI: 10.181103/mra.v11i11.4784.
[2] Singh A. Towards Early Prediction of Human iPSC Reprogramming Success. Machine Learning for Biomedical Imaging. 2023 Nov 11 DOI: https://doi.org/10.48550/arXiv.2305.14575.
[3] Zhao, Xy., Li, W., Lv, Z. et al. iPS cells produce viable mice through tetraploid complementation. Nature 461, 86–90 (2009). https://doi.org/10.1038/nature08267.
[4] Wenbo Zhou, Curt R. Freed, Adenoviral Gene Delivery Can Reprogram Human Fibroblasts to Induced Pluripotent Stem Cells, *Stem Cells*, Volume 27, Issue 11, November 2009, Pages 2667–2674, https://doi.org/10.1002/stem.201.
[5] These solutions should be portioned in 5mL aliquots, stored at -20C, and never frozen/thawed. Although antimycotics are unnecessary, CET highly recommends their usage for long-term cell culture. Antibiotics and antimycotics should not be used as an alternative to proper aseptic techniques.
[6] These solutions should be portioned in 5mL aliquots, stored at -20C, and never frozen/thawed. Although antimycotics are unnecessary, CET highly recommends their usage for long-term cell culture. Antibiotics and antimycotics should not be used as an alternative to proper aseptic techniques.

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